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CURRENT SERIAL RECORDS

GRAIN AND FORAGE CROPS RESEARCH

of the

United States Department of Agriculture  
and related work of the  
State Agricultural Experiment Stations

Section A

This progress report of U.S.D.A. and cooperative research is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on U.S.D.A. and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of U.S.D.A. and cooperative research issued during the last year. Current agricultural research findings are also published in the monthly U.S.D.A. publications, Agricultural Research, Agricultural Marketing, and The Farm Index.

UNITED STATES DEPARTMENT OF AGRICULTURE  
Washington, D. C.  
December 15, 1965





## ADVISORY COMMITTEES

The research program of the Department of Agriculture is reviewed annually by the following advisory committees:

1. Farm Resources and Facilities Research
2. Utilization Research and Development
3. Human Nutrition and Consumer Use Research
4. Marketing Research
5. Agricultural Economics Research
6. Forestry Research
7. Animal and Animal Products Research
8. Cotton Research
9. Grain and Forage Crops Research
10. Horticultural Crops Research
11. Oilseed, Peanut and Sugar Crops Research
12. Plant Science and Entomology Research
13. Tobacco Research

### ORGANIZATIONAL UNIT PROGRESS REPORTS

The source materials used by the advisory committees are of two types. First there are Organizational Unit Reports that cover the work of the Divisions or Services listed below. The number prefixes refer to advisory committees listed above that review all of the work of the respective Divisions or Services.

#### Agricultural Research Service (ARS)

- 1 - Agricultural Engineering
- 1 - Soil and Water Conservation
- 2 - Utilization -- Eastern
- 2 - Utilization -- Northern
- 2 - Utilization -- Southern
- 2 - Utilization -- Western
- 3 - Human Nutrition
- 3 - Clothing and Housing
- 3 - Consumer and Food Economics
- 4 - Market Quality
- 4 - Transportation and Facilities
- 7 - Animal Husbandry
- 7 - Animal Disease and Parasite
- 12 - Crops
- 12 - Entomology

#### Economic Research Service (ERS)

- 1, 5 - Economic Development
- 4, 5 - Marketing Economics
- 5 - Farm Production Economics
- 5 - Economic and Statistical Analysis
- 5 - Foreign Development and Trade
- 5 - Foreign Regional Analysis
- 5 - Natural Resource Economics
- 6 - Forest Service - Research (FS)
- 4, 5 - Farmer Cooperative Service (FCS)
- 4, 5 - Statistical Reporting Service (SRS)

The second type of report brings together the USDA program and progress for the following commodities and subjects:

- 6 - Forestry (other than Forest Service)
- 7 - Beef Cattle, Part I-a
- 7 - Dairy, Part I-b
- 7 - Poultry, Part I-c
- 7 - Sheep and Wool, Part I-d
- 7 - Swine, Part I-e
- 7 - Animal-Poultry and Products, Part II
- 8 - Cotton and Cottonseed
- 9 - Grain and Forage Crops
- 10 - Horticultural Crops
- 11 - Oilseed and Peanut
- 11 - Sugar
- 13 - Tobacco

A copy of any of the reports may be requested from W. C. Dachtler, Executive Secretary, Grain and Forage Crops Research Advisory Committee, Research Program Development and Evaluation Staff, U. S. Department of Agriculture, Washington, D. C. 20250

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## INTRODUCTION

This report on grain and forage crops research covers work directly related to the production, processing, distribution and consumption of grain, rice, feed, forage and seed and their products. It does not include extensive cross-commodity work, much of which is basic in character, which contributes to the solution of not only grain and forage crop problems but also to the problems of other commodities. Progress on cross-commodity work is found in the organizations' unit reports of the several divisions.

This report is organized by "Problem Areas" which are shown in the table of contents. For each area there is a statement of (1) the problem, (2) the USDA program, (3) State experiment station programs, (4) a summary of progress during the past year on USDA, and cooperative work, and (5) a list of publications resulting from USDA and cooperative work.

Research on grain and forage crop problems is supported by (1) Federal funds appropriated to the research agencies of the USDA, (2) Federal and State funds appropriated to the State agricultural experiment stations, and (3) private funds for research carried on in private laboratories or for support of State station and USDA work.

### Research by USDA

Farm research in the Agricultural Research Service comprises investigations on introduction, breeding and genetics, variety evaluation, culture, diseases, nematodes and weed control, insects, and crop handling and harvesting equipment and structures. This research is carried out in the Crops, Entomology, and Agricultural Engineering Research Divisions. It involves 359 professional man-years of scientific effort.

Nutrition, Consumer and Industrial Use research in the Agricultural Research Service pertains to composition and nutritive value, physiological availability of nutrients and their effects, new and improved methods of preparation, preservation and care in homes, eating establishments and institutions, new and improved food, feed, and industrial products and the processes related to grains, rice, and forages. It is carried out in the following research divisions: Northern, Southern, and Western Utilization; Human Nutrition; and Consumer and Food Economics. The work involves 280 professional man-years of scientific effort.

Marketing and Economic research is carried out in four Services. Grain, rice, feed, forage, and seed research in the Agricultural Research Service deals with physical and biological aspects of assembly, packaging, transporting, and storing, and distribution. It is carried out by the Market Quality and Transportation and Facilities Research Divisions. Work in the Economic Research Service deals with marketing costs, margins, and

efficiency; market potential; supply and demand; and outlook and situation. Consumer preference studies are carried out by the Statistical Reporting Service. Research on cooperative marketing is conducted by the Farmer Cooperative Service. The grain and forage research in these Services involves 56 professional man-years of scientific effort.

### Interrelationships Among Department, State and Private Research

A large part of the Department's research is cooperative with State experiment stations. Many Department employees are located at State stations and use laboratories and office space close to or furnished by the State. Cooperative work is jointly planned, frequently with representatives of the producers or industry affected participating. The nature of cooperation varies with each study. It is developed so as to fully utilize the personnel and other resources of the cooperators, which frequently includes resources contributed by the interested producers or industry. There is regular exchange of information between station and Department scientists to assure that the programs compliment each other and to eliminate unnecessary duplication.

Privately supported grain and forage crops research emphasizes the solution of specific production, processing, and marketing problems. Much of it utilizes the results of more basic work done by State stations and Department scientists. For example, private research is devoted to the synthesis and evaluation of chemicals as herbicides, fungicides, and insecticides, the development of new combinations of materials for use as fertilizers, product and process development, improvements in equipment for planting, cultivating, harvesting, handling, processing, drying and storing grains, forages and seeds. Private research in marketing and economics is largely concerned with studies of consumer preferences, market potential, promotion and market development.

The contributions of producers of grain and forage crops and of related industries to the work of State stations and the Department have been an important factor in the success of public research programs. Producers, processors, and distributors offer land, products, and facilities for the testing of equipment and practices used in the production and distribution of grain, rice, feed, forage, and seed and their products.

### Examples of Recent Research Accomplishments by USDA and Cooperating Scientists

Hybrid Barley Production Theoretically Possible. Commercial production of hybrid seed depends on a reliable and economical source of male sterile plants. This has prevented use of hybrids in crops in which cytoplasmic male sterility was not available or which could not be emasculated on a



large scale. In work with barley, a scheme has been devised that should overcome this problem for nonpolyploid crops. The method makes use of plants which have an extra interchanged chromosome that carries the linked genes for male sterility and a phytocide reaction located near the breakpoint. The male sterile gene insures a source of male sterile plants and the phytocide gene allows chemical roguing in the female rows to eliminate all fertile plants. Thus, seed production can be fully mechanized to keep the cost at a reasonable figure.

Improving Seedling Emergence in Intermediate Wheatgrass. Better stand establishment could be expected if varieties with improved emergence potential from deep seeding could be developed for range reseeding. Investigations with intermediate wheatgrass at Laramie, Wyo., suggest that good emergence is closely related to seed weight and coleoptile length. Analyses of diallel crosses indicated a strong maternal influence on the coleoptile length. However, long coleoptile females produced progeny with longer coleoptiles when crossed with long coleoptile males than with short coleoptile males. This suggests that improvement in coleoptile length, emergence potential, or seed size could be accomplished through simple maternal line selection with open-pollinated progenies. The effectiveness of selection based on seed size alone may be reduced by environmental effects.

Process for Reducing the Microbial Content of Flour. Department scientists have developed methods for treating wheats, as well as flours milled from them, to reduce the microbial population in the finished flours to very low levels--1 percent or less of the number present when the treatments are not used. The microorganism content of flours is important to the storage life of refrigerated, frozen, and cooked convenience foods. Therefore, specifications for the maximum permissible levels of bacteria, molds, and yeasts in flours are being included by most purchasers of flour for use in these convenience foods.

Without special treatments, flours are normally too high in microbial count for use in the convenience foods, despite the fact that 90-95 percent of the microorganism content of the wheat goes into the millfeed fraction. Our studies have shown that finished flours can be improved substantially by grain cleaning and the use of certain chemical agents in washing and tempering waters. However, when the level of microorganisms is high in the wheat, such treatments do not give a low enough count in the flour for critical uses. This difficulty has been overcome by developing processes based on using moderate heat treatment of the wheat before milling, or of the flour after milling. The finished flours have sufficiently low microbial count to meet specifications, yet show little or no heat damage to the flour.

Processes involving these heat treatments could be adapted to any mill with but moderate capital expenditures and with low operating costs.

Polysaccharide Gum from Corn Sugar Commercialized. Four industrial companies are now producing "B-1459" water-soluble gum by the Department's process for bacterial fermentation of corn sugar. Solutions of the gum in water exhibit unusually high viscosities that are stable over wide ranges of temperature and concentrations of salt, acids and alkali. In contrast, presently available gums are deficient in one or more of these qualities. Since Department feeding tests have shown negative toxicity, two of the companies have submitted requests for Food and Drug approval. The Department has assisted in commercialization of the gum by providing bacterial cultures and technical guidance and has cooperated with producers and possible users in evaluation of products and development of possible applications. The gum has a potential multimillion-pound market in industrial and food uses.

High Protein Rice Flours. Department scientists have shown that substantial quantities of rice flours of up to 16 to 20% protein can be abraded from the surfaces of either ordinary or parboiled milled rice. Currently available types of milling machinery are used, and thus the cost of producing the flour is quite low. If only three to five percent of the starting rice is milled off, breakage is negligible and the residual kernels are whiter and cook better. The color improvement is especially prominent with parboiled rice. The rice thus has a slightly higher sales value, perhaps high enough to offset the cost of producing the flour. The bland flours are non-allergenic, low in fiber, and provide high quality protein for infants, for adults with high blood pressure, and for elderly people who have dietary problems with normal sources of food protein. A large potential export outlet also exists to supply protein for weanling children in developing countries. The cooked flours from parboiled rice can be used directly to make gruels or they can be converted to powdered, soluble beverages in this country and then sold overseas. Further studies are being undertaken in cooperation with UNICEF and with industry to incorporate these flours into new products such as baby foods, beverages and soups.

Cooperative Work with Industry Provides Knowledge of Dehydrated Alfalfa Needed for Computing Least-Cost Feed Formulations. Department studies are leading to markedly improved feed materials at lower costs. The use of computers to calculate least-cost feed formulations has expanded greatly in the past several years. The proportions of each feed ingredient to be used are based on the relative costs of all the important nutrients available from competitive sources. Such complex calculations were impossible in pre-computer days. Precise analytical data for all nutrients in processed feed-stuffs is a prime requirement, but such data have not been generally available. Furthermore, adequate analytical methods for alfalfa products were lacking in some cases. To solve this problem, a cooperative basic research effort was undertaken by the Department's Western Utilization Division, two universities, a major feed company, a research institute, and the American Dehydrators Association. Dehydrated alfalfa from all major production areas of the country was subjected to exhaustive chemical analyses and



metabolizable energy assays. The first of a series of bulletins, containing only a part of the results, reached every segment of the industry within the first month after issue. Thus, an important gap in basic knowledge is being filled and this knowledge is being used by industry to make effective use of alfalfa meals in minimum cost feeds. The research is now being extended to other forage crops.

Wide Acceptance of Dryeration, a New Grain Drying Process. Dryeration, a combination of high-speed heated-air drying and aeration under development by Division engineers during the past two years, has gained wide acceptance. The process, in its first year of application was used to dry several million bushels of the 1964 corn crop. Cooperators report increases in drying capacity of from 40 to 80 percent, reductions in fuel and power costs, and cooler corn of better quality than they had experienced with conventional drying.

Bulk Feed Truck Operations and Cost. Preliminary findings in the study, including 110 bulk feed trucks operated by seven farmer cooperatives show: (1) Truck operating costs, not including administrative expenses, averaged 48 cents a mile operated, (2) operating costs averaged \$2.89 per ton of feed delivered, and (3) direct costs were over three-fourths of total operating costs.

Alfalfa Weevil Parasites Established and Spreading. The alfalfa weevil is continuing to spread in the eastern United States. Foreign parasite explorations have provided the importation of several species of parasites of this important introduced pest of alfalfa. Four of the species of parasites introduced are now established and spreading at one or more locations. Parasite releases have been made in Vermont, New York, Ohio, Indiana, and Missouri.



## I. FARM RESEARCH

### BARLEY CULTURE, BREEDING, DISEASE, & VARIETY EVALUATION Crops Research Division, ARS

Problem. Barley is subject to yearly fluctuations in yield and quality due to factors such as climate, soil, diseases, lodging, winterkilling, shattering, drought, insect damage, and other factors. Of immediate concern is the cereal leaf beetle which has appeared in Indiana, Michigan, and Ohio, and poses a threat to other grain-growing areas. Extensive screening tests are needed to locate genes for resistance to the cereal leaf beetle. Barley diseases continue to cause great losses in barley, to lower the quality of the grain, and to restrict the area where high quality barley can be grown. With the announcement of a new genetic system for producing hybrid barley seed, a great deal of additional basic work needs to be carried out to measure the degree of hybrid vigor (increased yield) that exists in this crop, on field management practices to insure a reasonable cost of the seed, on what effect hybrid vigor may have on malting and brewing quality, on the broad problems of disease and insect resistance, and on additional genetic studies to further perfect the new genetic system. Since this new system for producing hybrid seed is applicable to half of our cultivated crops, it seems logical that a maximum effort should be put forward now for barley as a test case in order to investigate the multiplicity of problems that arise with any new method. Commercial processing procedures for the production of malt and malt liquors are undergoing changes at the present time, and this raises the question of the suitability of presently used varieties for these purposes. The adequacy of the presently grown varieties needs to be determined, as well as to ascertain if other varieties may not prove superior with the new procedures. The genetics of resistance of barleys to aluminum toxicity in acid soils needs to be studied, and resistant varieties need to be developed for growing on such soils. Increased efforts are needed in genetics, cytology, pathology, physiology, and breeding methods to provide information basic to strengthen the breeding programs, the work on quality improvement, and to a fuller understanding of the nature of winterhardiness, host-pathogen relationships, and the relation to molecular structure to gene action.

### USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-range basic and applied program conducted by geneticists, pathologists, cytologists, physiologists, chemists, and agronomists, and related to the improvement of barley. Barley research investigations are conducted at Beltsville, Maryland, and in cooperation with the following State Agricultural Experiment Stations: Arizona, Idaho, California, Michigan, Minnesota, Montana, North Carolina, North Dakota, South Dakota, and Wisconsin. A Federal National Barley and Malt Laboratory is located at Madison, Wisconsin, and is operated in cooperation with the Wisconsin Agricultural Experiment Station. This Laboratory conducts quality research on barley for both State and Federal programs. The World Collection of barley varieties is maintained and distributed from Beltsville. Federal personnel supervise four spring and three winter uniform nurseries.

Five PL 480 projects dealing with research on barley are in operation in four foreign countries. In Egypt, project deals with studies on diseases affecting barley, namely, rusts, smut and net blotch. In Israel, on the screening and collection of cultivated barleys and related species, and breeding for resistance. In Poland, on finding sources of resistance to loose smut in barley and identifying genes for resistance. In India, on the physiological studies of drought resistance in some crop plants. The effect of genetic and environmental factors on the feeding value of barley proteins is being studied by Montana State College under a contract involving an estimated six-tenths professional man-year.

The Federal research effort devoted to barley totals 18.0 professional man-years, of which 9.6 is for breeding and genetics, 3.2 for diseases, 4.0 for variety quality evaluation, and 1.2 for culture (physiology).

#### PROGRAM OF STATE EXPERIMENT STATIONS

The States are engaged in both basic and applied research on barley. Most of them participate extensively in uniform regional tests and at many of the States, the research is conducted cooperatively with the Department.

Factors influencing the survival of winter barley and practices to enhance survival under severe winter environment are under study.

Plant breeders are developing improved varieties with higher levels of disease and insect resistance, better protection against lodging and winter-killing, and superior characteristics for malting or feeding purposes. The resources of material and information for breeding are being enlarged by genetic studies of factors for disease resistance in interspecific crosses, the effects of chemical and physical mutagens on genes and chromosomes, and the chromosomal location of various marker genes.

Diseases caused by fungi, nematodes, viruses, bacteria, or combinations of them are being studied. Emphasis is being placed on physiology of parasitism and mechanisms of pathogenesis. The chemical and physical detection of viruses in barley, their mode of action, means of transmission, strains, and mechanisms of survival are being studied. A number of foliar diseases caused by fungi, seedling diseases, and the role of barley crop residues in microbial interactions involving fungi and nematodes are under study. The role of nemas in the transmission of viruses is receiving increased attention.

Where barley is grown for malting purposes, the malting factors are evaluated to determine what effects various varieties and cultural practices and environments have on malting quality.

The total research effort on barley at 30 States is approximately 26.8 professional man-years, of which 1.0 is for culture, 18.1 for breeding and genetics, 4.8 for diseases, and 2.9 for variety quality evaluation.



## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Breeding and Genetics

1. Winter Barley Composite Cross. The hybrid seed from the male sterile composite cross produced in 1963 was increased in Arizona in 1963-64, and yielded about 850 pounds of F<sub>1</sub> seed. This breeding stock was released jointly by the Cornell and Arizona Agricultural Experiment Stations and the Crops Research Division, ARS, USDA. About 165 pounds of seed were distributed on request to breeders at 29 locations in the United States and in 10 foreign countries. A portion of the seed was planted in Arizona in which naturally crossed seed will be harvested from male sterile plants for further random recombination.

2. Hybrid Barley. A new scheme for producing hybrid seed of barley was announced during the year. It differs from all other schemes now in use and should work for all diploid plants - or about half of our cultivated plants. The scheme makes use of plants which have an extra interchanged chromosome that carries the linked genes for male sterility and a phytocide reaction located near the break-point. The male sterile gene insures a source of male sterile plants for the production of hybrid seed and the phytocide gene allows chemical roguing in the female rows to eliminate all fertile plants. Seed production can be fully mechanized in order to keep the cost at a reasonable figure. Early tests indicate that increased yields from hybrid seed will be in the range of 20 to 35 percent.

3. Frost Increases Natural Crossing in Barley. Light frosts at flowering time can destroy pollen, but not the egg in barley, thus inducing male sterility, and in turn out-crossing. At Davis, California, such a situation occurred, giving rise to 6 percent of natural out-crossing. Such events help explain the mass appearance of new genotypes in a breeding program and the persistence of heterozygotes in an otherwise self-pollinated crop.

4. Suppressor Gene Explains Reverse Mutation. A barley chlorophyll X-ray-induced mutant yv yellow virescent was re-irradiated, and near-normal green plants were obtained. Genetic studies involving original plants, the yv mutant, and the derived revertant disclosed that the reversion is due to a mutant recessive suppressor gene located at an independent locus. Chemical tests showed the yv mutant lacked chlorophyll-b, but that it was present in the derived revertant. These results indicate that the yv locus was not back-mutated and that the partial restoration of chlorophyll was due to the action of another gene.

5. New Variety Released. One new winter variety was released to seed growers. The two-rowed variety Tokak was released jointly by the New Mexico Agricultural Experiment Station and the Crops Research Division, ARS, USDA. Tokak was introduced from Turkey in 1949. Over a three-year period, under irrigation in the High Plains area of eastern New Mexico, Tokak yielded 11.6 bushels per acre more than the next best variety. Tokak is resistant to

lodging and shattering and has a high test weight. On dry land Tokak and other winter varieties may be damaged by winterkilling.

## B. Diseases

1. Micro Technique. Changes in plant cells induced by pathogens are being analyzed. A micro-culture technique for observing alterations in epidermal cells during the establishment and development of haustoria of the powdery mildew fungus has been developed. The breakdown of grana, stroma lamellae, and changes in osmophyllic granules in chloroplasts of cells invaded by the Barley Yellow Dwarf Virus (BYDV) are being followed by the use of an electron microscope.
2. Host Cell Physiology. Changes in host cell physiology resulting from the infection by pathogens are being studied. The effects of BYDV on the metabolic pathways within cells are being examined by enzyme analysis, C<sup>14</sup> labelled glucose, and paper chromatography. The powdery mildew fungus increased, but the stem rust fungus did not increase the respiration in tissue adjacent to that invaded by the fungal hyphae. The wheat powdery mildew fungus was propagated on barley plants infected with the barley powdery mildew fungus, but not on healthy barley plants.
3. Factors Affecting Virus Transmission. Factors related to infectivity and transmission of Barley Stripe Mosaic Virus (BSMV) are being determined. An inverse relationship between infectivity of BSMV and ribonuclease activity in stored, expressed plant sap has been shown. A higher percentage of plump seed transmitted the virus than thin or light seed. Two varieties, Moreval and Valmore, have been found that will transmit only a low percentage of the virus through the seed. Three genes conditioning the resistance of barley to the virus have been identified.
4. Intraspecific Fertility. The intraspecific fertility of 40 isolates of Helminthosporium sorokinianum, the fungus causing spot blotch, was not associated with geographic origin or original host from which the isolates were obtained. The H. sorokinianum isolates were incompatible with isolates of other H. spp.
5. PL 480 Project in Israel. Eight new physiologic races of the powdery mildew fungus which differed in pathogenicity from races previously identified in North America, Europe, and Argentina were identified. Mono-conidial isolates of the fungus were used to show that some varieties had unexpected genes conditioning their resistance to isolates of the fungus in Israel. Two new physiologic races of leaf rust which had not been reported previously were identified. Several collections of Hordeum made in fields in Israel have been found to be resistant to powdery mildew, leaf rust, and net blotch.

### C. Quality and Varietal Evaluation

1. Effect of Cultural Practices on Quality. In continuing studies at Madison, Wisconsin, spraying growing barleys with fungicides improved barley kernel color with no influence on quality. Barleys planted on April 16, 1965, at Madison, Wisconsin, were poorer in quality than those planted one week later. The second harvest date (July 26) produced the best quality. Swathing 2-row barley at Fort Collins, Colorado, at moisture levels above 42 percent and delaying swathing for 10 days reduced quality slightly.

2. Aleurone Layer of Cells Synthesizes Enzymes. The aleurone layer is the major site of enzyme synthesis in barley and the embryo supplies gibberellic acid type hormones. Modification of barley without embryo development in the presence of gibberellic acid was demonstrated in samples frozen at moisture levels of 30 to 40 percent. Enzyme synthesis and change of substrate constituents differed from those occurring during normal malting, but can be modified by conditions. More practical methods of inhibiting germ development without destruction of aleurone tissue have not been found to date.

3. Varieties from State and Federal Breeding Programs. More than 3,000 barleys from the 1963 crop were evaluated, approximately 2,500 by malting analysis. Fifty-nine advanced selections were carried through the brewing stage of evaluation, and 79 were processed through micro-brewing facilities. The new variety, Dickson, was equal to Trophy and Larker except for kernel size. Newer disease-resistant selections from North Dakota and Minnesota were not quite equal to the present high quality standards, Trophy and Larker. Two-row selections, excelling European introductions, are approaching release as varieties at several western Stations. In limited tests, the Upper Peninsula of Michigan produced good quality malting barley.

4. Emetic Compound. The emetic compounds in scabbed wheat are located in the outer layers of the kernel and were concentrated in the bran and tail-end short milling fractions. In Asiatic countries, scabbed grain could be reclaimed for human food by pearling. A Fusarium isolate obtained from Korea was identical to strains common here and produced emetic activity in artificial culture.

5. Malt Enzymes. The protease found in the largest quantity in malt was a hemoglobinase or possibly several closely related enzymes. A BAPA-ase, an amidase, and an esterase were also isolated and the first characterized more completely.

6. Amino Acids. The assimilation by yeast of amino acids from worts was influenced by total nitrogen level of the original barley, proteolysis during malting and pitching rate. Quantitative differences in individual amino acids were found in malt extracts from 10 varieties of barley. The origin and nature of differing peptides found in beer require further study.



#### D. Culture (Physiology)

1. Cell-Wall Polymers Related to Winterhardiness. Nonequilibrium freezing, which results in explosive formation of ice crystals, occurs in plants where a volume of liquid water in the space between living photoplasts has a uniform freezing point. In critical regions of winter cereals, the stress is greatly affected by substances which interfere with the development of the ice lattice and cause structural imperfections in the crystals. A polymer has been obtained from the crowns of hardened rye plants which was very effective as a competitive freezing inhibitor. Studies now are being directed to identify and characterize polymers in various plant species and varieties that are related to hardiness with the ultimate objective of determining to what degree polymer type and/or structure are under genetic control.

2. Differential tolerance of barley varieties to aluminum. Cooperative studies with the U.S. Soils Laboratory showed wide differences in winter barley varieties when grown on soils of low pH with a high soluble aluminum content. Smooth Awn 86, Dayton, and Colonial 2 were the most tolerant, and Kearney and Will were the least tolerant of 15 varieties tested in the greenhouse. Selections were made from the World Collection on a high aluminum soil and are being evaluated for possible use in breeding for aluminum tolerance. Crosses of tolerant and nontolerant varieties were increased in the F<sub>2</sub> generation for F<sub>3</sub> progeny tests to determine the method of inheritance.

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CORN AND SORGHUM CULTURE, BREEDING, DISEASE, AND VARIETY EVALUATION  
Crops Research Division, ARS

Problem. The hybrid seed industry has developed rapidly in recent years and now provides the major portion of the hybrid corn and sorghum seed sold. This development has permitted an extensive reorientation of the corn and sorghum program to give increasing emphasis to basic research in areas of genetics, physiology, and pathology. Progress is continuing but many problems still lack satisfactory answers. The phenomenon of heterosis on which the hybrid seed industry is based, still lacks a completely satisfactory explanation in either genetic or physiologic terms. Extensive work has been done on insect and disease resistance; but in many instances the mode of inheritance is still in doubt; and, in no case, has the biochemical basis of such resistance been established. Information is needed on the biochemical synthesis of oil, protein, and carbohydrates to increase the value of these crops from the standpoint of increased industrial or nutritional potential. Additional information is needed on resistance to corn viruses, on the potentialities of disease organisms to become infectious on currently resistant hosts, and to clarify the whole host-parasite interaction system.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving geneticists, physiologists, pathologists, and agronomists engaged in basic and applied studies relating to the improvement of corn and sorghum. Corn research is conducted at Tifton, Georgia; Beltsville, Maryland; Charleston, South Carolina; and Brookings, South Dakota; and in cooperation with State Experiment Stations at Tifton, Georgia; Urbana, Illinois; Lafayette, Indiana; Ames, Iowa; State College, Mississippi; Columbia, Missouri; Raleigh, North Carolina; Wooster, Ohio; Knoxville, Tennessee; and Madison, Wisconsin. Sorghum research is cooperative with State Experiment Stations at Manhattan and Hays, Kansas; Lincoln, Nebraska; Stillwater, Oklahoma; and Chillicothe and College Station, Texas. Research on improvement of corn, sorghum, and millet also is being conducted in Africa for the Agency for International Development.

Eighteen PL 480 projects dealing with corn and sorghum research are in operation. In India, projects deal with corn and sugarcane genetics; basic research on carbohydrate metabolism; the genetic effects of radiation; techniques to increase the storage life of pollen; storage, maintenance, and distribution of millets germ plasm; cataloguing and classifying genetic stocks of sorghums; responses of various germ plasm sources to agronomic practices; investigations on diseases of sorghums and other important millets; research on maize diseases with special reference to Erwinia carotovora var zeae; and studies on Helminthosporium species occurring on corn and sorghum. In the Philippines, research is conducted on downy mildew of corn; in Spain, on factors affecting the frequency of monoploid maize;

in Israel, on cytoplasmic sterility in plants; in Yugoslavia, on collection, classification, evaluation and preservation of local germ plasm; and on leaf redness in corn; and in Brazil, on evaluation of Latin American corn germ plasm for U.S. use.

The Federal research effort devoted to corn and sorghum totals 41.5 professional man-years. Of this, 25.7 are devoted to breeding and genetics; 8.2 to diseases, and 7.6 to culture and physiology. In addition, 5 professional scientists conduct research in Africa on corn, sorghum, and millet under funds provided by AID. Of these, 4 are devoted to breeding and genetics, and 1 to diseases.

#### PROGRAM OF STATE EXPERIMENT STATIONS

Scientists at the State Experiment Stations are engaged in basic and applied research in genetics, plant physiology, plant pathology, and agronomy. In many of the States, the research is conducted cooperatively with the Department. This research is continuing to provide useful fundamental information for the improvement of corn and sorghum production.

A number of States are determining the optimum plant population and spacing for specific hybrids and management practices for corn growing areas within the State. Other studies involve earless corn for silage and the effects of nutrient balance on corn diseases.

The management phases of sorghum receiving attention are date of seeding, plant population, number of harvests, irrigation, altitude adaptation, combined cropping with soybeans for silage, within-season cropping sequence with small grains and cover crops, and pollination factors in the production of hybrid seed.

In breeding of field corn most of the States are developing improved lines to fit local adaption or to meet local problems. The specific objectives of breeding are resistance to stalk lodging, drouth, corn borers, earworms, storage insects, stalk rot, leaf blight and other diseases, and higher oil, protein or amylose content. Attention is being given to the evaluation and improvement of breeding methods and the relative value of synthetics and hybrids. Other phases receiving attention are tetraploid types, sterility- and fertility-restoring factors, evaluation of foreign germ plasm, and interrelationships of Zea species.

Fundamental studies in the States concern population and biometrical genetics of corn, the effects of radiation on quantitative factors, characterization of genetic variation, mutation and paramutation systems within certain genotypes, and the genetics of host-parasite interactions of certain diseases. Increased attention is being given to characterization of corn stunt and maize dwarf mosaic diseases, and to a search for germ plasm resistant to these diseases.



A genetic factor conditioning cross-sterility between dent corn and popcorn is being studied and used in lines for popcorn hybrids.

Parental lines of sorghum for hybrids are being developed by States in the major sorghum-producing areas. The breeding objectives in these lines are resistance to lodging, drouth, diseases, insects and birds, seedling vigor, adaptation, improved head type and better grain quality. Texas is participating in a cooperative effort with the Department to convert short day-type foreign introductions into daylength types suited to United States environment.

Genetic studies are underway to determine species relationships in sorghum, to develop cytological stocks, to learn inheritance and linkage relationships of genetic factors, and to develop polyploids. Other studies concern the effects of radiation and colchicine for producing mutants and biometrical studies of the nature of gene action.

The quality of sorghum grain is being evaluated in feeding tests by cooperating animal science departments. The effects of cracking, grinding and pelleting of the grain on animal response are being determined.

The total research effort on corn and sorghum is approximately 108.5 professional man-years; of which 3.3 is for culture, 96.1 for breeding and genetics, and 9.1 for diseases.

#### A. Breeding and Genetics

1. Corn Genetics. Basic genetic studies are underway at Ames, Iowa; Beltsville, Maryland; Columbia, Missouri; and Raleigh, North Carolina. These studies involve three rather distinct areas: classical genetics and cytogenetics, quantitative genetics, and biochemical genetics. These have the common objective of a more complete understanding of the principles underlying the improvement of corn.

a. Classical Genetics and Cytogenetics. The unique conversion properties of the B allele appear not to involve infection, environmental induction or chromosomal aberrations. A particle or substance attached to the chromosome or an unusual property of the gene itself appears to offer the most likely explanation. Following infections with barley stripe mosaic, an unusual phenomenon has been discovered and designated "segregation distortion." Plant reciprocals involving affected individuals exhibit marked departures from the expected 1:1 segregations. Male and female transmission rates are essentially equal. Reversions may occur from high to low or from either high or low to normal. The permanence of the induced effect has been established, but no satisfactory explanation has been devised.

Studies on duplications of specific chromosome segments-- su, wx, and ae-- are underway to determine mode of chromosome pairing and the extent of duplications. Inheritance studies on "high-haploid" stocks are being continued. In studies conducted under a PL 480 grant neither silk nor pollen treatments have produced unequivocal evidence for an increase in frequency of haploidy. Stocks carrying an extra chromosome segment attached to chromosome 10, abnormal 10, are known to influence frequency of crossing over. Such material is being used to investigate the magnitude of linkage bias in estimates of genetic variance.

b. Quantitative Genetics. Four cycles of reciprocal recurrent selection have been completed involving two synthetic varieties, Stiff Stalk and Corn Borer No. 1. Yield increases have averaged approximately two percent per cycle. Crosses between the fourth cycle populations were comparable in yield to adapted double-cross checks. Comparisons of balanced sets of single and three-way crosses involving previously untested lines indicated the importance of epistasis. This is in agreement with similar studies involving lines selected for combining ability. Thus, interaction between loci is a factor of some significance in heterosis. Variance component estimates are being obtained in varieties of differing developmental background to provide information on the expected efficiency of different breeding systems. Quantitative genetic theory in the past has been largely limited to within varieties or populations. Theory is now being extended to inter-varietal hybrids to provide a more adequate basis for estimating genetic advance and for the interpretation of recurrent selection systems.

c. Biochemical Genetics. Types having approximately twice the normal concentration of xanthophyll have been developed to provide high-xanthophyll, high-energy feed for the broiler industry. A genetic study of carotenoid pigmentation is being continued to provide material for a biochemical study of synthesis. Types are now available which indicate genetic blocks at the phytoene and carotene-xanthophyll levels. Approximately 10 loci are known to affect the formation and character of anthocyanin pigmentation in the aleurone. Paired comparisons of developing aleurone tissue have given a consistent picture of the genetic blocks involved. Biochemical studies are underway to determine the specific compounds accumulating at each block.

## 2. Dent Corn Breeding.

a. Inbred Lines Released. Three inbred lines were released from the cooperative breeding program at Wooster, Ohio. These were Oh26F, Oh7K, and Oh422. The last two lines are resistant to maize dwarf mosaic.

b. High Xanthophyll Corn. High xanthophyll double-cross hybrids are now available which compare favorably with Dixie 82 in yield, lodging resistance, and resistance to the virus "corn stunt."

3. Sorghum Genetics. Genetic and breeding studies are underway at each of the locations where cooperative work is in progress. Cytogenetic studies are concentrated at College Station, Texas; disease resistance at Manhattan, Kansas and College Station, Texas; and the genetics of other traits at Hays and Manhattan, Kansas, and Lincoln, Nebraska.

a. Genetic and Cytogenetic Studies. Five distinct primary trisomics have been identified. Association of these with genetics marker stocks is being sought. A large number of additional trisomics are available and it is hoped the remaining 5 primary trisomics will be identified. Following irradiation, 30 plants carrying heterozygous translocations were identified. A series of translocations have been obtained in a homozygous condition and intercrossees are being evaluated to establish the identity of the translocated chromosomes. Mutations to resistance to milo disease caused by Periconia circinata have been observed repeatedly under conditions of heavy infestation.

Studies are underway to determine whether the toxin produced by the organism has mutagenic properties. To date mutations in the presence of the toxin have occurred at the rate of 1:3,300 and in the absence of toxin at the rate of 1:10,000. Numbers are not yet adequate to demonstrate significance for these differences. One of the height genes, dw<sub>3</sub>, appears to be mutable, with mutations from dwarf to tall. Comparisons have been made between reverted tall, DW<sub>3</sub> DW<sub>3</sub>, and the parental dwarf dw<sub>3</sub> dw<sub>3</sub>, forms. The tall type produced significantly higher yields and exhibited more heads per plant, heavier kernels, and a higher test weight.

4. Sorghum Breeding. Sterile and maintainer strains of K55, a forage sorghum line, were released in 1964 from the cooperative sorghum program at Manhattan, Kansas. Half of the approximately 1,000 elite lines from the World Sorghum Collection are currently involved in a backcross conversion program. This program involves the introduction into these strains, most of which will not flower in continental United States, of height and maturity genes which will make them potentially useful to United States sorghum breeders. The crosses are produced and F<sub>1</sub> hybrids grown at Mayaguez, Puerto Rico, and the F<sub>2</sub> generation at Chillicothe and Lubbock, Texas. Preliminary trials in 1964 indicate the feasibility of this approach. New sterile and restorer lines are under development and evaluation at Manhattan and Hays, Kansas, and Lincoln, Nebraska. In preliminary trials some of these appear superior to lines and hybrids now in commercial use.

## B. Diseases

### 1. Corn Diseases

a. Viruses. The most important corn diseases in 1964 were the two viruses, corn stunt and maize dwarf mosaic. Thus far these viruses are indistinguishable on the basis of symptoms but appear to have differing geographical distributions, host ranges, and methods of transmission. Corn stunt is confined to southern areas, has a limited host range, and is transmitted by leafhoppers. Two species of the genus *Dalbulus* can transmit the



disease. Probably other leafhoppers are also involved, although evidence on this point is still inconclusive. Many of the southern lines and hybrids appear to possess some resistance to this disease. Maize dwarf mosaic occurs to some extent in the southern States, but has caused its greatest damage in the Corn Belt. Losses in Ohio in 1964 due to this disease have been estimated at 5.8 million dollars. Maize dwarf mosaic is found in Johnsongrass and can be transmitted to corn, sorghum, sudangrass, and most of the grassy weeds infesting corn fields. Transmission is by mechanical means and by aphids. In the Corn Belt lines and hybrids tested thus far, resistance is rather infrequent. Comparative information is still inadequate, but it appears that two additional viruses have been identified. One causes some stunting, leaf discoloration, and red striping on the pericarp. This virus occurs in the northwestern Ohio, northern Indiana, and in Michigan. This virus can be readily transmitted to wheat. The second virus has been reported from Iowa. It can be transmitted to certain broadleaved hosts and in this respect, is unique.

b. Corn Rusts. Studies involving Puccinia polysora indicate eight races. In each case resistance appears to be due to two genes. Present evidence suggests that at least one member of each pair of genes for resistance is linked. Results from a continuation of this study should have an important bearing on host parasite relations.

c. Downy Mildew. Downy mildew (Sclerophthora) on corn and sorghum has been observed as far north as Stillwater, Oklahoma. Information on resistance is largely lacking. A PL 480 project in the Philippines has demonstrated that types resistant to Sclerospora can be isolated.

d. Helminthosporium. Work on this genus has been directed toward an understanding of pathogen population structure and the potentiality of a pathogenic organism to undergo changes in host specificity and pathogenicity. Extensive collections of Helminthosporium species reveal 5 rather distinct types with respect to conidial morphology. Intratype crosses exhibit fertility ranging from 55 to 80 percent, while intertype fertility averaged 0.29 percent. Studies on pathogenic abilities indicate that most populations within types have a number of common genes for pathogenicity. Intertype crosses, however, provide a mechanism for gene transfer and, thus, increase the genetic potential for changes in pathogenicity. In crosses involving H. victoreae, there is a correlation between toxin production and pathogenicity crosses involving isolates of H. turcicum, nonpathogenic to selected lines of corn, may give rise to new strains which are highly pathogenic to one or more of the previously resistant lines. Breeding for disease resistance must, therefore, recognize the genetic potential for modification of both parasite and host.

2. Sorghum Diseases. The most serious diseases of sorghums in the Great Plains area are smuts and charcoal rots.

a. Head Smut. Losses up to 40 percent have been observed in commercial fields in the Coastal Bend region of Texas. Losses in Kansas were estimated at 200,000 bushels in 1964. Sources of resistance to head smut are now available, and the incorporation of such resistance is progressing rapidly.

b. Charcoal Rot. The incidence of charcoal rot is erratic but under favorable conditions may cause extensive losses. Conditions favorable for infection include (1) plants in milk to dough stage; (2) daily maximum temperatures above 35° C.; and (3) a sub-lethal moisture stress. Summer temperatures in Arizona are favorable for the development of the disease, and water stress can be regulated through control of irrigation water. A contract has been negotiated with the Arizona Agricultural Experiment Station to screen inbred lines for resistance. Additional refinements in inoculation techniques are required, but preliminary observations indicate genetic variability for susceptibility to water stress and to susceptibility to charcoal rot.

c. Other Diseases. Two types of downy mildew, Sclerophthora macrospora and Sclerospora sorghi, appear to be on the increase. Some nursery materials grown at College Station, Texas in 1964 were completely susceptible. Another disease, tentatively designated "stunt," produces symptoms similar to Sclerophthora. Oospores, however, are lacking and mechanical and aphid transmission to sorghum, Chenipodium and Nicotiana, have been possible. These results indicate the causal agent is a virus. This disease was first observed in 1957 and has been recorded in each succeeding year.

## C. Physiology and Culture

### 1. Corn

a. Protein Metabolism. Three different RNase's have been isolated from corn kernels. These are the soluble RNase A, the particle bound RNase B, and the RNase M, which is found on microsomes. It was established that Illinois High Protein corn is low in RNase while Illinois Low Protein corn is high in RNase. Strains having intermediate protein percentages are currently being evaluated. Current results would suggest that RNase may exhibit a direct control in protein synthesis, possibly through the destruction of messenger RNA. Protein bodies are a characteristic feature of the corn endosperm. Protein bodies were isolated and tested for their ability to incorporate amino acids. The results were quite inconclusive as all incorporation could be accounted for as a result of bacterial contamination. Techniques have been developed to reduce contamination and the studies will be repeated with freshly harvested endosperm tissue.



b. Carbohydrate Metabolism. Normal corn starch is composed of approximately 75 percent amylopectin and 25 percent amylose. Genetic mutants are known which modify this ratio; e.g., waxy (wx) which produces approximately 100 percent amylopectin while amylose extender (ae) and dull (du) in combination produce high percentages of amylose. It is known that when a reaction involving synthesis is blocked, either by an inhibitor or by a mutant, the intermediates preceding the block tend to accumulate. It appears feasible through use of several mutants affecting carbohydrate reserves or their combinations, to obtain information on the pathway of starch synthesis. A number of enzymes presumed to play a role in starch synthesis have been isolated from the corn kernel. The exact role of each remains unclear. The enzyme, starch synthetase, in the presence of a suitable primer and ADP-glucose can produce starch. This enzyme, however, is lacking or in very low concentration in waxy (wx) types which produce normal amounts of starch. This suggests that alternative pathways may be involved.

c. Mineral Nutrition. Inbred lines of corn are known to respond differentially under conditions of magnesium deficiency. Oh40B exhibits magnesium deficiency symptoms under conditions where WF9 remains normal. When these two lines are grown hydroponically at various Mg levels, the leaves of Oh40B are low in Mg and exhibit typical deficiency symptoms. Differences in Mg concentration between Oh40B and WF9 were not apparent in either roots or stems. Thus, deficiency symptoms appear to be related to limitations on translocation rather than absorption. An increase of Mg in the nutrient solution reduced the K, P, Ca and Fe content of both lines. A continuation of studies of this type should provide important information on the role of the mineral elements in plant nutrition and on their interactions.

2. Sorghum. A cooperative program on sorghum physiology was initiated at Lincoln, Nebraska, in June of 1964. Initial emphasis will be placed on photosynthesis and translocation. This will be supplemented by studies on water stress, providing rapid and precise methods for estimating the free energy status of internal water can be developed. A contract has been negotiated with Nebraska to develop and evaluate a microwave refractometer to estimate internal water activity.

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WHEAT AND RYE CULTURE, BREEDING, DISEASE, AND QUALITY INVESTIGATIONS  
Crops Research Division, ARS

Problem. Wheat is grown commercially in 42 States, and to some extent, in five others; rye is grown in 31 States with sparing use made of it in some others. This obvious diversity in cultural environment is accompanied by a very diverse range of problems of adaptation, disease and insect pests, and of utility of the crop, including milling and baking quality. The problem, therefore, is to develop suitable practices, improved varieties, and better pest controls, to maintain or improve the quality of the crop produced, and to develop new knowledge as a basis for further improvements. This requires a broad range of disciplines in the scientists who conduct the work, and it requires a number of locations in order to be situated where the specific work can be done most effectively.

USDA AND COOPERATIVE PROGRAM

The Department has a long-term program of research leadership in wheat and rye investigations. The objectives of the research are to stabilize production by reducing losses, to increase efficiency of production, to improve the quality of wheat and rye wherever grown in the United States, and to accumulate and disseminate knowledge. Basic and applied research is performed in the laboratory, greenhouse or field, and involves the sciences of agronomy, physics, and statistics. Regional and national leadership is given to several phases of the work.

The World Collection of Wheat Varieties is maintained and distributed from Beltsville, Maryland.

Work is conducted on 15 diseases of wheat to obtain basic information which helps to improve methods of control. This requires the attention of 20 professional employees. Milling and baking quality evaluation utilizes 20 professional employees in checking new varieties and conducting basic research on mechanisms involved in quality measurements. Hybrid wheat work, basic genetics, germ plasm development, and breeding new varieties involve 19 professional workers. Culture and physiology problems are on a small scale with only 1 professional worker-equivalent devoted to this phase of work; it involves stand establishment and plant hardiness.

There is no manpower on rye. Some work is included at a few locations that, combined, amounts to less than one-half man-year of effort.

PL 480 projects on wheat and rye include the following: In Spain, polyploidy in rye, and cereal rusts; in Egypt, cereal rusts and smuts; in Pakistan, cereal rusts and smuts; in Poland, pathogenic diversity in rust; in Israel, root systems, and origin of cereals; in India, 3 rusts of wheat.



Federal effort is concentrated in eight research centers. Additional locations are required either to provide proximity to the problem or to take advantage of facilities or contacts with workers located at such locations. There are 49.6 professional man-years assigned to this area, of which 17.9 are for breeding and genetics, 18.5 for diseases, 12.0 for variety evaluation, and 1.2 for culture and physiology.

#### PROGRAM OF THE STATE EXPERIMENT STATIONS

Scientists at the State Experiment Stations are engaged in basic and applied research in plant breeding and genetics, plant pathology, agronomy, cereal technology, and starch and protein chemistry. In many of the States, the research is conducted cooperatively with the Department. This research is continuing to provide useful fundamental information for the improvement of wheat production.

Studies on production practices concern rates and dates of planting and rates of fertilization as they interact with specific varieties and their effects on milling and baking quality. The merits of various types of tillage implements for seedbed preparation are being compared.

Many States are involved in wheat breeding with each devoting attention to the market class or classes grown within the State. The major objectives of wheat breeding are resistance to the rusts and smuts, virus diseases, greenbug, Hessian fly, and winterhardiness in the winter varieties. Improved milling and baking qualities and shorter stiffer straw receive high priority in many of the current breeding programs. Plant introductions are being studied for useful characteristics. Work related to the development of hybrid wheat involves a search for sources of cytoplasmic male sterility and fertility-restoring genes, the transfer of sterility and restorer systems into breeding lines, procedures for hybrid seed production, tests for first and later generation hybrid performance, and the development of lines for possible use in hybrids.

Genetic studies on wheat involve the inheritance of genes for insect and disease resistance, host-pathogenetic relationships, the development of monosomic series of wheat lines, studies of chromosome irregularities, and a search for genetically controlled reactions to herbicides or chemicals. Cooperative work at Missouri concerns the evolution of wheat, interrelationship of species and genera, the effects of chromosome V on chromosome pairing, and the study of centromere structure.

Variety evaluation for quality and various phases of cereal chemistry are receiving attention in 12 States. The four Federal regional wheat quality laboratories are located in four of these States and cooperate closely with the State researchers. Much effort is devoted to the evaluation of varieties for quality as well as the effects on quality caused by various environmental factors during the growing season, conditioning of grain before milling, and crop management factors. A new regional project NCM-41 is being initiated; and under it, the biochemical and physical characteristics of the wheat

kernel and its components such as protein, starches, lipids, enzymes, and pigments will be studied for their influence on wheat quality. Weather, storage conditions, milling and processing practices will be studied as factors causing changes in quality of wheat. Fractionation of flour, based on particle size and density, with air-classification is being studied as well as the role of various enzymes in wheat flour.

The total research effort on wheat and rye is approximately 47.3 professional man-years; of which 1.4 is for culture, 32.0 for breeding and genetics, 13.4 for diseases, and 0.5 for variety quality evaluation.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Breeding and Genetics

1. New Varieties and Germplasm Released. 'Chris' was released for growing in the northern spring wheat States. It is a hard red spring wheat with good quality and unusually high resistance to stem rust (cooperative with Minnesota).

'Lewis' was released in Missouri as a shortstraw, productive, soft red winter variety.

'Stadler' also released in Missouri, has good performance including high resistance to leaf rust. Both Lewis and Stadler originated from irradiated breeding stocks.

'WRT 238-5' is a germ plasm release of a wheat x rye translocation stock. Seed in the fourth backcross to Pembina, Justin, Selkirk, and other varieties is available. The very high type of stem rust resistance comes from rye. Normality was noted except the translocation gametes were not transmitted through pollen equally. A virescent strain was also established from this material, but it is poorly transmitted (cooperative with Missouri and Minnesota).

2. New Wheats in Advanced Tests. New and very promising selections include semidwarf hard spring wheats in Minnesota combining good yield with standability and disease resistance, semidwarf hard red winter selections in Texas with special promise for irrigated land, stripe rust-resistant club wheats in Oregon and Washington, stripe rust-resistant Lemhi and Thatcher types in Idaho and Utah, a sawfly-resistant winter wheat in Montana, and a sawfly-resistant spring wheat in North Dakota. Seed of these is under preliminary increase.

3. Hybrid Wheat Progress Slowed by New Problems. Two serious problems met with in hybrid wheat this year involved full sterilization of the male-sterile parent and the counterpart of full fertility restoration. A number of varieties possess one major gene, and others have certain minor genes for fertility making it difficult to stabilize sterility in them under diverse environments. Cheyenne, Selkirk, Itana--to name only 3--have partial restoration genes. Environment was noted to alter expression drastically. Light intensity may be the key factor in restoration stability. Over 200 varieties are in the process

of being converted by backcrossing to male sterile forms. Far fewer restorer varieties are being developed because the process is slower and intervening test crosses must be conducted.

4. Agropyron and Aegilops. Wheat-like derivatives transmit high leaf rust, streak mosaic, and stripe rust resistance. Streak mosaic resistance appears to be conditioned by one chromosome. Irradiation-derived stocks of the latter are under test in Oklahoma. If successful, badly needed stocks to combat this disease will be obtained.

5. Rye and Wheat Chromosomes Related. Rye chromosomes were shown to have homoeology with wheat groups 2,3,4, and 6. Plants with the substituted chromosomes are reasonably vigorous and fertile. This research will help in bridging species barriers.

6. Aluminum Tolerance. Tolerance to acid soils containing high levels of KCL-extractable aluminum was shown to be an inherited characteristic in wheat and was demonstrated to be an important element in variety adaptation (cooperative with SWC). Root growth was affected as much as the tops. Tolerant varieties included three introductions from Brazil, Seneca from Ohio, and Atlas from North Carolina.

7. Natural Evolution in Rye. Rye is a major field and roadside weed in northern mountain counties in California and adjacent Nevada and Idaho. Great diversity is evident in this material indicating evolutionary changes might be obtained for adaptation factors.

8. Dalapon Mutants. Ramona wheat yielded types shorter, later, and more stripe rust resistant when plants were treated with dalapon.

9. Red Pericarp Color Gene of Red Bobs Found on Chromosome 3A. Monosomes were utilized to locate the genes. Earlier, a gene was found on chromosome 3B from P.I. 178383 that influenced color (Oregon) and, in another study (Missouri), chromosome 3D was involved. Now with the discovery of a gene on 3A, all factors of the R<sub>1</sub>, R<sub>2</sub>, R<sub>3</sub> series have been located and correspond to the chromosome 3 homoeologs. These are valuable genetic markers.

10. Red Glumes and the T Gene Linked. Bunt resistance (T factor) and a gene conditioning red glumes are on chromosome 1B, about 14 units apart. The M<sub>1</sub> factor for resistance appeared to be independent. These data help to clarify the genetic information on bunt resistance. In this case, results from a cross of Columbia X Elgin supported earlier monosomic data that genes R, which imparts the same resistance as T, was located on 1B; but it is still uncertain whether the R and T factors are the same. Tests are underway to determine if an inversion is involved.

11. Three Genes were Noted Influencing Hybrid Necrosis. Many varieties, when intercrossed, give F<sub>1</sub> plants that die as seedlings or persist weakly. This genetic barrier must be overcome or circumvented to produce hybrid seed either for conventional breeding programs or for commercial hybrid wheat. At least



three genes are known to condition this response. In a special experiment, T. macha, Prelude, and Triumph were studied. The Ne<sub>3</sub> factor was tentatively located on chromosome 3D.

12. CCC Responses. Culm dwarfing by 2-chloroethyl-trimethylammonium chloride spray was found to be modified by genes located on chromosomes 6A and 2B in a cross of Lemhi<sup>2</sup> monosomes X P.I. 178383. The main effects are delayed internode elongation, delayed heading, longer period of flowering, and increased tillering. The utility of these responses for hybrid wheat seed production has not been explored.

13. Two of the Known 3 Genes for Bunt Resistance from the Highly Resistant P.I. 178383 have been Isolated. A third gene is tentatively isolated. Curiously, all three isolated lines are susceptible to bunt race D-6, and this suggests that a fourth gene is present in P.I. 178383. This P.I. variety resists all known races of bunt in the United States, both common and dwarf forms.

14. Cereal Leaf Beetle Feeding Trials. In Michigan and Indiana, tests revealed over 500 wheats with promise for a retest to confirm resistance. A preponderance of these were noted to have pubescent leaves (cooperative with ENT), but physiologic mechanisms may be involved also since some nonpubescent varieties either escaped or resisted feeding by the insect. Crosses utilizing the most resistant varieties have been made to combine suitable adaptation and market properties with resistance.

15. Introgression from Aegilops into Wheat. An important contribution to the evolution of modern forms of Triticum was traced according to recent research in Israel on a PL 480 project. Genome homologies were shown to be strong in test crosses of Aegilops with Triticum. Evidence also was obtained on chromosomal modification in the evolutionary process.

16. Two Rye Varieties Produced in Georgia by Mass Selection are Weser and Emory. Weser resists leaf rust and has expressed some tolerance to anthracnose. Emory, selected from Explorer, has general disease resistance and good adaptation in all of Georgia. Anthracnose has seriously curtailed the usefulness of rye, especially where it would be desirable to sow consecutive crops on the same field. While Weser is better than most other varieties, it, too, may be damaged, so higher levels of resistance are being sought.

17. New Tetraploids of U.S. Rye. Stocks were found to be stable in two advanced generations. These are now entering polycross nurseries for studies of cross-compatibility and heterosis (Spain, PL 480 project E25-CR-5). Colchicine was used to double the chromosome numbers in the varieties.



## B. Diseases

1. Soilborne Wheat Mosaic Virus Infection Closely Timed. The effect of time and temperature on inoculation of wheat has been determined. Debris from soil collected from the field was good inoculum Sept. 15-March, but poor collected April to Sept. 15. The winter wheat plant is in a juvenile stage during the period when inoculum is most abundant. These studies narrow the time when inoculation probably occurs.
2. Wheat Streak Mosaic Virus Concentration. It appears highest in the younger fully expanded leaves of a seedling plant grown at 20-21° C. A serum is being sought for serology uses. Analytical density gradient centrifugation is being used to determine virus concentration. The best source yielded 10-20 ug of virus from one gram of leaves.
3. Cellular Disorders Were Revealed. Electron microscope studies on wheat tissues invaded by wheat streak mosaic virus and barley yellow dwarf virus show abnormal starch grains and cell bodies. The chloroplasts are one of the first and most severely affected organelles in the cell and such disruption markedly affects the physiology of the cell. Research is continuing on breakdown of cell membranes and possibly altered enzyme systems.
4. Stem Rust Infection Reduced Roots. Growth and respiration rate and fewer roots contribute to early maturity of diseased plants. Wheat roots apparently have little food reserves and are dependent upon continuous supplies from the shoots. Rust causes a decline in the normal flow of carbohydrates to roots; and with heavy infection, root starvation occurs leading to premature death of the plant. The rust treatments were supplemented by studies using steam to simulate the effect of rust. One-inch portions of basal stem tissue were steam-killed at a time corresponding to rust sporulation.
5. Respiratory Rate at the Center of Rust Colonies. The rate was 15 times that of healthy tissue. This grades off completely to normal at the margin of lesions. Mildew gave different results in that effects extended beyond hyphal growth.
6. A Culture of Race 15 Attacks Wells and Lakota. Also cultures of races 11, 32, 38 and 87 attacked key varieties in trace amounts. In addition to greenhouse tests, these cultures were used in variety studies in Puerto Rico. Resistance to all of the cultures was noted among germ plasm sources in the trials.
7. Elite Groups of Broadly Rust-Resistant Varieties have been Assembled. Nonspecific resistance as well as other mechanisms are represented. From 367 varieties evaluated, 22 had resistance to a culture of race 32B, which is pathogenic on Selkirk, Crim and Justin. In a species collection, 138 entries had broad seedling resistance and 58 additional ones showed resistance in the mature plant stage. Tolerance levels, while difficult to establish and define, were evident.

8. Slower Rates of Rust Epidemic Development. . Chemical treatments result in a reduction of the inoculum potential. Spores trapped within isolated plots are proportional to the incidence of disease in that plot.
9. Spores were Collected in Rain Water Before Being Detected on Slides. Rain-deposited spores caused infection. Suggested hypothesis is that these may be primarily involved in epidemic spread.
10. A Type Culture Collection of Plant Rust Fungi Established. Success with liquid nitrogen storage of rust spores has led to the possibility of a widely based type culture collection for genetic and pathology research.
11. Thirty-Six Different Pathogenic Cultures. This pattern has been noted for stripe rust on 12 differential varieties in Washington. No culture yet attacks Bersee, Alba, Ministre, or P.I. 178383.
12. Stripe Rust Genetic Studies. Gaines in 13 crosses indicated that simple genes were involved; in 7 crosses there were 2 genes. In 4 crosses of Omar, one gene was indicated. These genes are not all the same one or two. P.I. 178383 seems to carry one dominant gene giving a broad spectrum of resistance, and minor genes.
13. International Rust Nursery. Now in its 15th year of operation, it is effective in testing about 1,000 new accessions annually to rust cultures in some 25 countries on 6 continents. It has up-graded the quality and effectiveness of wheat breeding in many parts of the world including the U.S.A.
14. Mildew Tests Involving 36 Cultures and 280 Varieties were Completed. Only 'Khapli' resisted all cultures. However, a number of highly resistant varieties were noted.
15. Fifteen Varieties of Hard Red Winter and Soft Red Winter Were Examined for Blackpoint Disease on the Seed. Helminthosporium and Alternaria fungi were the most often isolated. Eleven fungi were tested for root rotting.
16. Calcium Pantothenate Increased Rust Infection. The number of plants becoming infected with stem rust and the number of pustules per plant were increased. Adding this chemical to hypodermic suspensions should increase rust inoculation efficiency. A concentration of  $10^{-2}M$  was used.
17. Pathogenic Specialization in Dwarf Bunt Confirmed the Existence of Two Additional Dwarf Bunt Races. Race D-9, which has been collected in several localities in southeast Idaho, attacks all of the major bunt resistance factors including Hohenheimer. Race D-10, which is represented by collections from Blind Springs, Utah, and Missoula, Montana, attacks the R<sub>idit</sub> and M<sub>2</sub> factors but is avirulent on the M and T factors. These studies suggested also that modification of the dwarf bunt race classification may be required with regard to the reaction of certain races on the Hussar and Hohenheimer factors.

18. Varietal Resistance to Dwarf Bunt. None of the entries deriving resistance from the commonly used bunt resistance factors exhibited adequate levels of resistance to races D-3 and D-9. However, several PI selections and entries representing crosses with P.I. 178383 exhibited complete or nearly complete resistance to these most broadly pathogenic bunt races.
19. Dwarf Bunt Infection Closely Correlated with the Stage of Plant Development. Plants that were just beginning to tiller at the onset of the infection period (January-March) were the most susceptible to infection and produced the highest number of smutted heads. Laboratory studies indicated that germination of dwarf bunt spores can occur and can persist over an extended period of time at temperatures slightly below freezing, provided the spores receive a preceding period of incubation at temperatures above freezing.
20. Phleum Boehmeri Shown to be Susceptible to Dwarf Bunt. Infection of Phleum adds a new genus to the host list of this fungus.
21. The Incidence of Smut Presented a Double Image. Common smut was again negligible as a production factor, extending to six years the period of its relative unimportance in the Pacific Northwest. Dwarf smut, on the other hand, was a major problem in some of the known areas of infestations. In Flathead Valley around Kalispell, Montana, it was devastating, some fields exhibiting upwards of 75 percent smutted heads. At least one field was estimated at over 95 percent smut.
22. Dwarf Bunt Discovered in Commercial Fields of Gaines. This was reported from the Moscow-Genesee area of northern Idaho and the Rosalia-Rockford area of southeastern Washington. These are essentially contiguous areas and there is a long history of dwarf bunt in the general locality, although it has not been a serious production factor in perhaps 10 years.
23. Mating Plus and Minus Lines of 19 Bunt Races. Infection patterns have resulted which indicate that multiple sex groups do exist in Tilletia caries and T. Foetida.
24. Approximately Half of the 84 Winter Wheat Entries Submitted for Bunt Testing in 1964 had a Level of Resistance Equivalent to Either Omar, Burt, or Gaines. The successful transfer of the resistance of P.I. 178383, which is immune to all races of common and dwarf bunt, was evident in 15 entries.
25. Only One Pathogenic Race of Powdery Mildew Present in the Pullman, Washington area. None of the currently popular commercial varieties exhibit good resistance to the existing powdery mildew. However, there is good mildew resistance in a number of selections being used in the advanced breeding program, one of which is P.I. 178383.



26. Results from Seed-Treatment Tests. TCNA (2,3,5,6-tetrachloronitroanisole) has provided good control of both the seedborne and soilborne phases of common bunt. The effective rate of application was 1 ounce of a 50 percent formulation per bushel of seed. The control provided was comparable with that obtained with HCB (hexachlorobenzene). In addition to HCB and TCNA, two other formulations gave good control of soilborne bunt in tests conducted in 1964. These materials were: Hercules 3944 Seed Treatment Formulation x and Chemagro 4797.

27. Dwarf Bunt Control was Obtained Near Kalispell, Montana. Soil surface applications of HCB, TCNA and PCNB (pentachloronitrobenzene) were used. Results showed minimum rates of application for the formulations used to be 5 pounds per acre for HCB and TCNA and 2-1/2 pounds per acre for PCNB. However, on an active ingredient basis all three chemicals were similarly effective. The HCB and TCNA formulations contained 40 and 50 percent active ingredients, respectively, while the PCNB formulation was 75 percent wettable powder.

28. Field and Greenhouse Evaluation for Flag Smut Resistance. The commonly grown varieties Gaines and Burt, as well as the Gaines Sib referred to as Burt 7, all have similar reactions to flag smut. About 30 percent flag smut developed in these varieties in the greenhouse, but lower percentages occurred in the field. Several excellent sources of flag smut resistance were detected and notable among the resistant wheats was P.I. 178383, which had no smut in this test. New centers of flag smut were reported near Athena, Oregon; Pullman and Dusty, Washington.

#### C. Variety Evaluation for Quality

1. Pigments and Lipids in Durum and Other Classes. Solvents have been found but better ones are needed to separate and purify the extracts. At least 6 carotenoid-like compounds were extracted. Lipids restrict and mask the separations. Lipids from single wheat kernels were separated by thin layer chromatography into 15-17 components.

2. A 2-Speed Farinograph Procedure for Testing of Dough Reveals New Rheological Properties. The high speed (126 rpm) is equally good for general mixing properties and absorption but gets results in half the time. The slow speed (53 rpm) was better for time and tolerance tests.

3. Poor Pastry Quality Resulted from Wheat Grown on Sulfur-Deficient Soil. At high yield levels on one soil in Idaho, omitting gypsum from the fertilizer brought out marked deficiencies in flour milling behavior and in baked products. The flour yield was several percent lower and the ash content of the flour was 40% higher than from wheat grown on properly fertilized plots. Cookies had poorer spread and top grain from sulfur-deficient flour.

4. Very Soft Kernels Were Separated from Club Wheat and the 2 Lots Compared. The milling excellence normally seen in Club wheat was evident in both lots. It was concluded that chalkiness and mottling of club wheat kernels was no indication of different milling behavior in such samples.



5. Gaines Mills as Well as Most and Better than Some Western Wheat Varieties Marketed as Soft White. Omar Club is superior in milling even when test weight per bushel is one or two pounds below that of common soft white varieties.
6. Gaines, Brevor, and Omar Quality Compared from 6 Crop Harvests from 4 States. All three were equally satisfactory as pastry flours and made good cookies and cakes. Gaines and Brevor were superior to Omar in cake-baking tests. Omar had lower viscosity than the other varieties which is desired for limited market use. In these trials, Gaines had lower protein content.
7. Cake Volume Shows High Relationship with Particle Size Index and Break Flour Yield. The fracturing characteristics of wheat endosperm appears associated with the cake-baking potential of a variety. This discovery may lead to an improved procedure for predicting quality in soft wheat.
8. Starch Separation with 95 Percent Efficiency was Achieved by a New Procedure. Hard as well as soft wheats were processed.
9. Using Crystalline Amylases, Beta-Amylase Contributed at Least 10 Times More Fermentable Sugars than Equal Amounts of Bacterial, Pancreatic, or Fungal Amylases. Mixtures of alpha and beta forms yielded less sugars than calculated.
10. Pure Proteases from 6 Sources Compared. Profoundly different effects on loaf volume were noted over the same range of hemoglobin units of potency. Dosage rates of certain sources caused a marked change in dough-mixing properties.
11. Low and High Quality Wheats were Fractionated. Protein that was recovered was further separated into lactic acid insoluble gluten, gluten soluble at pH 6.1, and that precipitated at 6.1. Gluten soluble at pH 6.1 was inversely related to loaf volume and mixing time.
12. Alveogram Studies. It was shown that the length of curve and loaf volume gave the best correlation when the optimum bromate level was used.
13. Changes in Lipid Composition During Wheat Development were Followed. Qualitative and Quantitative thin layer chromatography (TLC) and fractionation on silicic acid columns were used. Development of the wheats was accompanied by a slight decrease of lipid content on an as-is basis, and by almost doubling of lipids on a kernel basis. Free fatty acids in mature wheat were less than half the amount in 21-23 days preripec wheat. A similar decrease was found in the levels of mono- and di-glycerides. No consistent changes were found in phospholipid fractions of lipids extracted from wheat at various stages of maturity. Carotenoid pigments disappeared as the wheat kernel matured and developed. Deterioration of fully mature wheat, however, results in markedly reduced amounts of phospholipids and disappearance of several of them. This breakdown was more rapid than formation of free fatty acids. Varieties were found to differ in lipid composition.

14. Dispersibility of Wheat Flour Proteins in 3M Urea in 0.05 M Pyrophosphate Buffer pH 7.0 at 4° C. The results were correlated with bread-baking potentialities of samples representing 6 hard red winter wheat varieties from 19 locations and of samples representing composites of hard red winter, northern spring, soft red winter, durum, and soft white (Club) varieties from 8 locations. Urea-dispersible proteins, expressed on an equal protein basis, had consistently higher absorbence readings at 280 mu in poor than in good bread-baking wheat flours. Absorbence of urea-dispersible proteins of flour increased as a result of deleterious effects of -irradiation of wheat. The protein dispersibility index was negatively correlated with bread-baking potentialities of flours milled from the wheats. It is postulated that ease of dispersibility and disaggregation of proteins in urea is related to lability and unfolding of wheat proteins during mechanical- or heat-processing of wheat dough and might explain large differences in oxidation requirements of flours which vary little in levels of total sulfur- or thiol-containing amino acids.
  15. Proteins were Extracted with a 3M Urea Solution at 4° C. The amino acid composition of the urea-extracted protein contained upon hydrolysis higher amounts of ammonia than a commercial soya protein isolate. Amino acid analysis by ion exchange chromatography showed that on an ammonia-free basis, the urea-extracted proteins were comparable to a commercial soya protein extract; and both had an amino acid composition comparable to that of a 50 percent protein commercial solvent-extracted soya meal. The urea-extracted proteins from wheat flour and soya flour retained their functional properties in breadmaking.
  16. Evaluation Tests Made on 2,200 Samples Submitted by Plant Breeders for Milling and Baking, and 867 for Macaroni Studies. In addition, single property or dual-property tests were conducted on approximately 30,000 samples.
- D. Culture and Physiology
1. Defoliation Reduced Protein Content. Standard and high-protein lines were similarly affected when the leaves were removed during kernel enlargement and filling. The flag leaf had the most effect.
  2. Certain Gibberellins Alter Dormancy Patterns. GA<sub>3</sub> and GA<sub>4</sub> reduced post-harvest seed dormancy in 4 varieties. Dwarf plants from some crosses which normally do not head-out in the greenhouse were induced to head when treated with GA<sub>3</sub> and grown at high temperatures. These treatments accelerate the research schedule and permit studies on some genotypes that formerly were lost.
  3. Height Studies Show That Yield and the Semidwarf Habit are not Necessarily Related. The high yield obtained from Norin 10 (seen in Gaines) was transferred to tall genotypes. Conversely, this means that short straw is not in itself a yield factor and that other plant characteristics must be bred into the high yielding genotypes. However, short stature may permit the genotype fuller opportunity to be expressed.

4. Temperature-Sensitive Stem Rust Reaction Associated with the Sr6 Gene Studied for Light-Temperature Interaction. Incandescent and fluorescent light gave equal results. During the cool treatment studies light could be introduced either in the warm or cool cycle and so long as a minimum daily exposure to low temperature was provided (rather than a continuous exposure) resistance was expressed.
5. Fluoroacetone Fungicide Proved in Reducing Damage from Stripe Rust in Oregon Tests. Some phytotoxicity was noted. In stem rust and leaf rust studies in Minnesota, this material was not as effective as nickel plus maneb and was more phytotoxic.
6. Subcrown Internode Length. This character and coleoptile length and plant height are correlated. The proper balance among these, crown depth, and hardness is important for best adaptation.

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OAT AND BUCKWHEAT CULTURE, BREEDING, DISEASE, AND VARIETY EVALUATION  
Crops Research Division, ARS

Problem. Most of the United States oat crop is produced under warm, humid conditions highly favorable for disease development. The major limiting factors for profitable oat production in the United States have been a constant acute need for new disease-resistant varieties. Other problems are lack of adequate straw strength, adaptation, nutritive value, winter hardiness, yield, and grain and forage quality.

Buckwheat has been long neglected from the standpoint of breeding for increased yield, grain quality, straw strength, disease resistance, adaptation, etc.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term research program involving pathologists, geneticists, and agronomists engaged in both basic and applied research in an attempt to solve the problems besetting the oat crop. The ultimate objective is to stabilize production, increase efficiency, and improve the quality of oat grain and forage wherever it is grown. Most of the work on the breeding, genetics, and disease of oats is conducted in cooperation with State Experiment Stations in Arizona, California, Florida, Georgia, Idaho, Illinois, Indiana, Iowa, Kansas, Minnesota, Mississippi, Missouri, New York, Pennsylvania, South Dakota, Texas, Virginia, Washington, and Wisconsin; the Puerto Rico Agricultural Experiment Station; and at Beltsville, Maryland. Oat research is being conducted under three PL 480 projects. One in Poland on crown rust of oats; a second in Colombia, S.A., on extremely virulent races of oat stem rust; and the third in Israel on collecting and screening wild species of oats for resistance to virulent races of stem and crown rust.

Buckwheat breeding and genetics are limited to an industry-supported graduate student at University Park, Pennsylvania.

The Federal scientific effort devoted to research in this area totals 13.7 professional man-years. Of this number, 5.8 is devoted to breeding and genetics, and 7.9 to diseases.

PROGRAM OF STATE EXPERIMENT STATIONS

Scientists at the State Experiment Stations are engaged in basic and applied research in plant breeding and genetics, plant pathology and agronomy. In many of the states, the research is conducted cooperatively with the Department. This research is continuing to provide useful fundamental information for the improvement of oat production.



A regional project of the northeastern region, NE-23, deals with the improvement of oats for resistance to winterkilling and to barley yellow dwarf virus (BYDV). The project is concerned with environmental and genetic factors influencing the development of winterhardiness. The studies of BYDV concern the properties and the variability of the virus and the biology of the cereal aphids as related to their role as vectors of the virus. Federal personnel of the Department located at New York and Pennsylvania cooperate on this project.

Work in other parts of the country involves breeding for local adaption plus stiff nonlodging straw and resistance to diseases and insects. The diseases of greatest concern are BYDV, rusts, smuts, Victoria blight, and septoria; and the insects receiving most attention are greenbugs or aphids. Cold resistance is being studied from the standpoint of greater winter grazing in the North and greater winterhardiness in the South. The action of mutagenic agents acting separately or in combination; interspecific crosses; inheritance of factors for disease resistance; interrelationships of seed characteristics and yield; and geometry of yield are studies underway to provide information, materials, and techniques for more efficient varietal improvement programs.

The total research effort on oats is approximately 28.6 professional man-years; of which 1.4 is for culture, 23.4 for breeding and genetics, and 3.8 for diseases.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Breeding and Genetics

1. Improved Oat Varieties. Tippecanoe is an unusually stiff-strawed, high-yielding, rust- and smut-resistant, spring oat variety developed cooperatively by the Purdue University Experiment Station and ARS. Its straw strength is much better than that of either of the parents of the cross, Clintland 60<sup>2</sup> x Mo. 0-205, from which it was selected.

Santee is a short, stiff-strawed, early-maturing, high-yielding, spring oat variety developed cooperatively by the Nebraska Agricultural Experiment Station and ARS. Its primary area of adaptation is northeastern Nebraska, where it has yields comparable to the high-yielding, later-maturing varieties.

Pennfield is an exceptionally stiff-strawed, high-yielding, but relatively low-test-weight, late-maturing spring oat variety developed cooperatively by the Pennsylvania and Idaho Experiment Stations and ARS. It responds especially well to high fertility and early seeding. Pennfield is resistant to many of the prevalent races of rust and smut.

Houston is a short-statured, high-yielding, and high-test-weight winter oat with near spring-type growth habit, developed cooperatively by the Texas Agricultural Experiment Station and ARS. It has greater lodging resistance than any other oat variety grown in Texas. Houston is an early forage producer and is resistant to Victoria blight, culm rot, and most prevalent races of crown rust.

2. Irradiation of Oats. The Texas Agricultural Experiment Station, in cooperation with ARS, has concluded a ten-year study on the effect of X- and neutron-irradiation of five oat varieties. In addition to the expected chlorophyll mutants and influence on viability and vitality, irradiation apparently was responsible for significant changes in reaction to low temperatures, Victoria blight, and specific races of crown rust. The new Alamo-X was one of several superior agronomic lines selected from this irradiated material.

3. Oat Monosomes. Six monosomes produced through X-irradiation of Cherokee oats have been designated A,B,C,D,E, and F, by the Iowa Station and ARS. The L (Landhafer) gene controlling reaction to crown rust and a gene controlling seed fluorescence are located on monosome D. Monosome F can be recognized by its fatuoid character. Several genes for chlorophyll deficiencies have been located among the six monosomes. Genes A,B, and D, controlling reaction to stem rust, gene V, controlling reaction to crown rust and Victoria blight, and a gene for pubescent leaf sheath are not located on monosomes A,B,C,D, or E.

4. Inheritance of Morphologic Characters in Oats. The mode of inheritance of lemma color, awns, shape of primary floret base, type of floret separation, floret basal pubescence, rachilla hairs, panicle shape, and numerous aberrant types have been studied in the F<sub>2</sub> and/or F<sub>3</sub> generation of some 40 oat hybrids. With few exceptions these characters showed mono-di and tri-hybrid segregation with varying degrees of dominance.

5. New Genes from Wild Species. An intensive program of collecting and testing for rust resistance among wild species of *Avena* is being conducted in Israel under a PL 480 project. Sources of resistance to highly virulent U.S. races of crown and stem rust have been found in Israel within the diploid, tetraploid, and hexaploid species. Preliminary data indicate that these probably new genes are, in general, resistant to specific races or groups of races but not to a broad spectrum of races. The resistance being found in the hexaploid *Avena sterilis* is of special interest since genes available in this species should be readily transferable to cultivated oats. The high degree of variability for disease reaction, plant height, vigor, maturity, growth habit, pubescence, grain size and color, and other characters observed among the *A. sterilis* collections would indicate this species should afford a rich source of new genes for oat improvement.

6. World Oat Collection. During 1964 the World Oat Collection received 101 selections from 18 states and 4 countries, and 405 samples of six species of *Avena* from 6 foreign countries. The oat collection now contains approximately 8,000 entries. The Small Grain Collection prepared and shipped 13,871 samples of oats to 45 plant scientists located in 18 states and 13 foreign countries.

7. Cereal Leaf-Beetle Resistance. In cooperation with Entomology Research Division and Michigan Agricultural Experiment Station, 4,738 entries of cultivated oats in the World Oat Collection were exposed to cereal leaf beetle oviposition and feeding. None of the entries were free of damage, 1,158 showed moderate damage, and 3,363 were severely damaged. Some of the entries in the "trace" and "moderate" feeding damage classes apparently escaped damage as a result of growth habit. The apparently moderately resistant entries of cultivated oats are being re-tested along with available entries of wild species of *Avena*.

## B. Diseases

1. Identification of the Barley Yellow Dwarf Virus (BYDV) Particle. In cooperation with the New York and Nebraska Agricultural Experiment Stations, BYDV was purified from frozen BYDV-infected oats by differential centrifugation following clarification by chloroform. Infectivity assays were based on feeding aphid vectors through membranes on test inocula. The BYDV particle is a dense polyhedral about 30 mu in diameter. The particles were photographed and measured by use of an electron microscope. The BYDV particles were obtained from viruliferous aphids and their pathogenicity verified.

2. Sources of Resistance to Barley Yellow Dwarf Virus (BYDV) in Oats. More than 10,000 oat varieties, selections, and species were tested in the field and greenhouse for reaction to BYDV in cooperation with the Illinois Experiment Station. Thirteen selections of the wild diploid *Avena strigosa* and 13 cultivated hexaploid varieties and selections were rated as tolerant to four strains of BYDV; whereas, 450 entries were rated moderately susceptible, and the remaining 9,500 entries as either highly or extremely susceptible. The named varieties Albion (C.I. No's. 729 and 1050) and Fulghum (C.I. No's. 1951 and 1962) appeared to represent the best and possibly different sources of hexaploid resistance. The diploid Saia (*Avena strigosa*) (C.I. No's. 4639, 6954, and 7010) appears to represent the highest type of protection from BYDV currently available in oats.

3. An Intermediate Reaction to Victorin. Victorin, the specific pathotoxin produced by *Helminthosporium victoriae*, is the disease-inducing entity of Victoria blight. Susceptible reaction to the toxin is controlled by a single dominant gene. A spontaneous mutant (BRM 280) of Fulgrain oats exhibited an intermediate sensitivity to victorin compared with the susceptible variety Fulgrain and a resistant mutant BRM 281. Permeability, as measured by rate of loss of electrolytes from victorin-treated tissues, and relative root growth were used to measure reaction to the toxin. The intermediate mutant



also has an intermediate Victoria-type reaction to certain key races of crown rust. The mutant is thought to be the result of a selection of part of the "Victoria-locus."

4. Helminthosporium Victoriae Compared with H. Sorokinianum. Four isolates of the victorin-toxin-producing H. victoriae and six of H. sorokinianum were selected for a detailed morphological and physiological study. Two of the H. sorokinianum isolates produced typical culm rot symptoms on susceptible oat varieties. Comparisons of conidia shape, color, and measurements; and rate of mycelial growth under different temperature and pH showed that the four isolates of H. victoriae made up one homogeneous group and the six of H. sorokinianum made up a second fairly homogeneous group. The culm-rotting isolates of H. sorokinianum has some mycelial growth characteristics similar to H. victoriae but were morphologically similar to H. sorokinianum and were likewise nontoxin producing. It was concluded that H. victoriae should be retained as a separate species and that the culm rot fungus should be classified as H. sorokinianum.

5. Effect of Light and Temperature on Reaction to Oat Stem Rust. Research studies under a PL 480 project in Colombia and in cooperation with the Iowa and Minnesota Agricultural Experiment Stations have revealed varying degrees of sensitivity to light and temperature during incubation of certain genes in oats controlling seedling reaction to specific races of stem rust. Of the genes A,B,D,E, and F, controlling reaction to oat stem rust, only gene A appears not to be affected by either or both varying light intensity or temperature. The resistance afforded by genes D and F is lessened with a change from low to high light intensity, and the resistance of B,E, and F is lessened with a change from low to high temperature. The unidentified gene or genes controlling stem rust resistance in the diploid Saia were affected by light but not temperature.

6. International Register of Races of Crown Rust. Crown rust race numbers 201 to 347 have been assigned, and their reactions to 10 standard differential oat varieties recorded, in cooperation with the Iowa Agricultural Experiment Station. A key to facilitate rapid determination of unknown races has been published. Scientists in 11 countries cooperated in the assignment of race numbers.

7. Races of Oat Smut in the Southeastern States. Ustilago avenae is the predominant oat smut species in the southeastern United States. Race A-22 has been the most prevalent and widely distributed race of U. avenae in this area in recent years. Atlantic and Fulghum, among the host tester varieties, were resistant to A-22. Two Victoria-attacking races (A-20 and A-23) were widely distributed but not as prevalent as A-22. A new race (K-9) of U. kolleri was collected for the first time near Vicksburg, Mississippi. Adequate sources of resistance are available to all of the races identified.



## C. Variety Evaluation

1. Increased Winter Survival in Winter Oats. The variety Winter Turf has long been considered to be the most winter hardy oat variety. This has not been true in recent years. A summary of winter survival data from uniform winter oat hardiness nurseries for the past 36 years reveals that 40 percent of the 342 entries tested during 2 or more years of this period exceeded Winter Turf in average survival. The top five named varieties for winter hardiness during the past 10 years were Wintok, Ballard, Excel, Nysel, and Norline, with a range in average survival of 143 to 131 percent of Winter Turf. The 10 best experimental selections for the same period ranged from 150 to 141 percent of Winter Turf.

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RICE CULTURE, BREEDING, DISEASES, AND VARIETAL EVALUATION  
Crops Research Division, ARS

Problem. Numerous problems in rice production are created by diverse demands of processors and consumers; the wide range of soil types, cultural methods, and climatic conditions; and the varying prevalence of diseases and insects. Varieties are needed which are adapted for each area and resistant to pests and other environmental hazards. Varieties also must have proper milling, cooking, and processing characteristics to satisfy changing consumer or processor requirements. Changes in cultural practices have created new problems. The increased rate of application of nitrogen fertilizer is associated with higher losses by the blast and kernel smut disease and more lodging. Low temperature of the irrigation water in California has created a serious problem in stand establishment.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program that involves plant breeders, geneticists, plant pathologists, and chemists who are engaged in basic and applied research relating to the improvement of varieties and the control of diseases. Rice research is conducted at Beltsville, Maryland; and in cooperation with State Experiment Stations at Stuttgart, Arkansas; Biggs, California; Baton Rouge and Crowley, Louisiana; and Beaumont, Texas. The work at Biggs, California, is also in cooperation with the California Cooperative Rice Research Foundation, Inc.; and at Beaumont, Texas, with the Texas Rice Improvement Association, and private enterprises interested in rice quality.

The project with the Japanese Ministry of Agriculture and Forestry on the blast disease which was initiated in 1963 under the U.S.-Japan Scientific Corporation Program, was continued during the year.

Rice research also is conducted under four PL 480 projects: One with the Central Rice Research Institute, Cuttack, India, on the nature and variability of resistance of rice to the blast fungus and in physiological specialization in the causal organism; one with the Institute of Botany, Academia Sinica, Taipei, Taiwan, on cytogenetics of rice and its related species; one with the Taiwan Agricultural Research Institute, Taipei, on physiological races of rice blast fungus; and one with the Taiwan Provincial Chung-Hsing University, Taichung, on genetic studies of mutations induced by radiation.

The Federal scientific effort devoted to research in this area totals 8 professional man-years. This number includes 5.0 devoted to breeding and genetics; 2.0 to diseases; and 1.0 to quality evaluation.



## PROGRAM OF STATE EXPERIMENT STATIONS

Scientists at the State Experiment Stations are engaged in basic and applied research in genetics, plant physiology, plant pathology, and agronomy. Most of the research is conducted cooperatively with the Department. This research is continuing to provide useful fundamental information for the improvement of rice production.

Studies on production practices deal with land preparation for water and dry seeding, water management during the growing season, the use of chemicals for weed control, organic residues and rotation programs. Field practices are also being evaluated for their effects on quality.

Breeding is being done for local adaptation on long, medium, and short grain types. Resistance to hoja blanca and blast are being sought along with improved plant types. Genetics studies are underway on chromosomal structure to determine relationship between species and a possible connection with sterility in interspecific crosses. Other genetic studies concern the mode of inheritance of endosperm characteristics and the development of aneuploid lines.

The total research effort on rice is approximately 7.2 professional man-years, of which 0.0 is for culture, 3.9 for breeding and genetics, and 3.3 for diseases.

### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

#### A. Breeding and Genetics

1. New Variety Release. A new variety, 'Bluebelle', developed in the cooperative breeding program in Texas, was released for production in 1965. Bluebelle (C.I. 9544) is a lodging resistant, smooth-hulled, very early maturing, long-grain variety. Compared with 'Belle Patna', the new variety is superior in "first crop" production, has shorter and stiffer straw, is about a week later in maturity, and the grain is similar in type and cooking characteristics. Seed enough for about 1,200 acres was distributed.
2. World Rice Collection. Varieties in the World Collection have been grown and classified for several morphological characters and length of growing period; grain type; chemical and physical properties of the grain; and reaction to blast, hoja blanca, and straighthead. About 1,000 varieties were grown in 1963, and over 3,000 in 1964, to complete this work which has been underway for several years. The collection contains a wide diversity of types, some of which will be useful in breeding programs.

A total of 128 new introductions were grown at Beltsville in 1964, and the seed produced was sent to field stations for growing in 1965.

3. Plant characters associated with straw strength in rice. F<sub>2</sub> populations from crosses of varieties that differed in unit culm weight, culm diameter, and breaking strength of culm and the relationships among these characters were studied. Differences in breaking strength of the culm were much higher than for unit culm weight and culm diameter. Unit culm weight and culm diameter had only a minor influence on culm breaking strength so there are one or more additional culm characters that influence straw strength.

4. Breeding high protein rices. The protein content of progeny (F<sub>8</sub>) from crosses of high and low protein varieties and the parent varieties was studied. In one cross the two parent varieties had protein contents of 8.00 and 10.75 percent, respectively. The protein content of progeny from the cross of these two varieties ranged from 8.36 to 11.79 percent. The protein content of the parent varieties and the progeny vary from year to year, but the "high" protein varieties are always comparatively high and the "low" varieties comparatively low. There was a correlation of -.57 between grain yield and protein content among progeny of one cross. However, some lines with relatively high protein content also produced relatively high grain yields.

5. Inheritance and relationship to yield and height of four morphological characters of rice. The inheritance and relationship to plant height and yield of pubescence, presence or absence of ligules, length of outer glume and awnedness were studied. Pubescence was dominant to glabrous and liguled was dominant to liguleless with a single gene difference in each case. Long outer glume was recessive in one cross and semidominant in another cross. Awnedness was dominant to awnless with a single gene difference in the cross where long outer glume was recessive. There was no correlation between plant height and yield within crosses. Ligules strains significantly outyielded liguleless strains in both crosses studied. Short outer glume strains were significantly higher in yield than long glume in the cross where long glume was semidominant but not in the other cross. Presence or absence of awns was not associated with yield. No combination of characters outyielded the preferred combination of awnless, ligules, short glume, and glabrous.

6. Cytogenetic studies of rice and its related species. This research was in Taiwan under a PL 480 research project. In one study, Oryza sativa and O. officinalis were crossed and the F<sub>1</sub> was backcrossed to O. sativa. Seeds obtained from the backcross produced triploid plants. This was explained on the basis of the union of a haploid O. sativa gamete and a diploid gamete from the O. sativa X O. officinalis hybrid.

7. Seedling vigor in rice. Seedling vigor is an important character in rice. Varieties that germinate in a short time and grow rapidly during the seedling stage often emerge to a stand; whereas, less vigorous varieties may not if unfavorable conditions prevail. Seedling vigor was studied under controlled conditions. A method was developed which gave reproducible results. This method is useful in evaluating seedling vigor of varieties and breeding lines.

## B. Diseases

1. Blast. All United States rice varieties are susceptible to one or more races of the rice blast fungus (Piricularia oryzae). However, sources of resistance are available to each race known to occur in the United States.

A study of the mode of inheritance of reaction to race 1 and race 6 of P. oryzae showed that resistance was dominant and monogenic in each case and that genes Pi<sub>1</sub> and Pi<sub>6</sub> were not linked.

In the blast research program under the PL 480 project in India, 16 races of P. oryzae were identified. Two of these were similar to U.S. races 8 and 16.

2. Hoja blanca. No rice plants with hoja blanca were found in the United States in 1964.

The reaction to hoja blanca of 18 rice varieties was studied in the field at Palmira, Colombia, during the 3-year period 1961-53. The prevalence and severity of hoja blanca were determined by (1) visual rating on a scale of 0 to 9; (2) counting the number of diseased plants; and (3) counting the number of diseased tillers. There was good agreement in the rating by the three methods. On the basis of the results obtained, the 18 varieties were divided into four classes - susceptible, moderately susceptible, moderately resistant, and resistant. The reaction of varieties was similar from year to year, and the reaction of varieties was in agreement with the results obtained in previous tests in other countries. These facts indicate that only one strain of the hoja blanca virus was present or dominant in Colombia during the years of this experiment or in other Latin American countries prior to 1961.

## C. Variety Evaluation for Quality

1. Physical and chemical characteristics of commercial rice varieties and advanced breeding lines. Commercial varieties and advanced breeding lines of the long- and medium-grain types were evaluated using tests that characterize the cooking and processing properties of rice. Rice produced at four locations in the Uniform Yield Nurseries was used. The tests were conducted to determine the iodine blue value, alkali reaction, protein (Biuret) value, gelatinization (transition) temperature, and other amylograph values, water uptake at 77° C., and dry matter loss when parboiled. The results from these tests gave information that is essential for deciding upon the release for commercial production of the breeding lines.



#### D. Culture and Physiology

1. Influence of light intensity and nitrogen fertilizer on yield and yield components. The effect of three levels of light intensity and three levels of nitrogen fertilizer on rice yield and components of yield were studied at four locations for 3 years. It was found that increased light intensity increased the utilization of nitrogen resulting in increased yield, and reduced light during early growth stage had less adverse affect on yield than reduced light during the tillering and flowering stages. The principal components of yield were found to be seedling stand, seeds per panicle, panicles per unit area, and panicle weight.

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ALFALFA CULTURE, BREEDING AND  
GENETICS, DISEASES, AND VARIETAL EVALUATION  
Crops Research Division, ARS

Problem: More than 28 million acres of alfalfa are grown annually in the U. S. under a wide range of environmental conditions. Climatic and pest hazards subject the crop to great fluctuations in yield and quality. Substantial increase in basic and applied research is needed to stabilize production and to cope with presently recognized hazards. For example, spread of the alfalfa weevil seriously threatens alfalfa production in the eastern and western States. Varieties resistant to this and other pests are urgently needed. Some problems are not readily solved by breeding, and their solution depends on management or other measures. Practical solutions rely on a continuing program of basic research.

USDA AND COOPERATIVE PROGRAM

The research program includes: Basic breeding and genetic studies; breeding for resistance to alfalfa weevil, potato leafhopper, spotted alfalfa aphid, pea aphid, bacterial wilt, common leafspot, and Leptosphaerulina leafspot; basic studies on pathogens causing disease; and cultural and physiological studies on cutting management, diseases, and temperature stress. Research is conducted at: University Park, Pa., Ithaca, N. Y., Beltsville, Md., Raleigh, N. C., St. Paul, Minn., Brookings, S. D., Lincoln, Nebr., Manhattan, Kans., Logan, Utah, Reno, Nev., and Stoneville, Miss. Research on resistance to pests was strengthened at University Park, Pa., Beltsville, Md., Raleigh, N. C., and Reno, Nev. All work is in cooperation with the respective State Agricultural Experiment Stations, except at Beltsville, Md. Most entomological research is in cooperation with the Entomology Research Division.

There is a P.L. 480 contract at Perugia, Italy, to develop lines of alfalfa resistant to Verticillium wilt.

The Federal scientific effort devoted to research in this area totals 15.6 professional man-years. Of this number, 9.7 are devoted to breeding and genetics, 2.8 to diseases, and 3.1 to culture and physiology.

PROGRAM OF STATE EXPERIMENT STATIONS

Scientists at the State Experiment Stations are engaged in basic and applied research relating to alfalfa breeding and improvement. These scientists include geneticists, pathologists, physiologists, entomologists, and agronomists. Much of the research is coordinated on a regional basis and conducted cooperatively with the Department.



Regional research projects NE-28, Forage Crop Breeding, and NE-29, Forage Crop Management, along with other related regional efforts, have aided materially in the coordination and progress of alfalfa research.

Since the ultimate objective of this program is to produce a quality forage for efficient production of animal products, a great deal of attention has been focused on the search for superior varieties and strains to meet this objective. This requires close contact with the animal scientists and the identification of guidelines for directing breeding programs. More specific objectives for alfalfa improvement include (1) breeding for disease and insect resistance, (2) development of varieties and strains for adaptation to specific soils and climates, (3) better information on physiology relating to heat, cold, and drought tolerance, and (4) better understanding of cultural practices which might influence stand, yield, and quality.

In research at the Stations, scientists are learning more of the mechanisms of resistance and the discrete processes of virulence of the causal agent of bacterial wilt of alfalfa. Microbial assays now in progress promise to provide much needed information on the nematodal and fungal diseases of roots. Recent findings through the characterization of cellulolytic and pectolytic bacterial populations of the alfalfa rhizosphere provide a new approach to controlling certain root pathogens. Recent discoveries on the role of the calcium ion on enzymes produced by various isolates of root pathogens of alfalfa offer much promise in the eventual control of these organisms. Studies in progress on the respiration and metabolic pathways in organisms such as Phymatotrichum omnivorum, will also eventually make possible their control. The use of germ-free tissue culture systems is providing needed knowledge on nematode pathogens of alfalfa.

The total research program of the States involves approximately 49.3 professional man-years of which 11.5 is for culture, 24.9 for breeding and genetics, 10.5 for diseases, and 2.4 for variety evaluation.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Breeding and Genetics

1. Root-knot nematode resistance. Winter-hardy alfalfa lines selected at Reno, Nev., were resistant to northern root-knot nematode (Meloidogyne hapla) in Nevada, Oregon, Utah, and Washington. Parent plants simplex for resistance have been intercrossed and duplex resistant progenies recovered.

2. Resistance to common leafspot reduces coumestrol content. Research at Brookings, S. D., and University Park, Pa., showed that 3 cycles of selection for resistance to Pseudopeziza medicaginis within each of 2

pools of alfalfa germplasm increased leafspot resistance and reduced levels of the estrogen coumestrol. Leaflets with no lesions contained no coumestrol; whereas, leaflets with 10 to 20 lesions averaged 85.9 ppm in pool A and 178.8 ppm in pool B.

3. Delta alfalfa. A new alfalfa variety, Delta, will be released in 1965 in cooperation with the Mississippi Agricultural Experiment Station. Delta alfalfa planted in heavy clay soil in the Yazoo-Mississippi Delta area of Mississippi remained productive for 5 years. This appeared to be due in part to its tolerance to root and crown rots, leafhopper yellowing, and certain leafspot diseases. Limited quantities of certified seed of Delta are expected in seed trade channels by fall 1966 or 1967.

4. Saponin content. At Logan, Utah, a correlation coefficient of  $+0.28^{**}$  was obtained between chalcid infestation and saponin content on 62 alfalfa varieties.

5. Alfalfa weevil resistance. Field and laboratory studies showed that 70 clones selected at Beltsville, Md., differed significantly in numbers of egg masses deposited per stem, numbers of larvae per stem and larval feeding damage. Oviposition accounted for approximately 18 percent of the variation in larval feeding damage. Eggs deposited per stem and punctures per stem were highly correlated, indicating that number of egg masses was a suitable measurement of oviposition. An association between small stem size and resistance to egg laying was demonstrated. Characteristics of Medicago falcata tended to be associated with resistance. At Raleigh, N. C., finding that  $BC_1S_1$  progenies from the (gaetula x Cherokee) x Cherokee cross had improved levels of resistance indicated progress from breeding.

6. Temperature alters heritability of resistance to spotted alfalfa aphid. At Manhattan, Kansas, heritability of nymph survival calculated from the regression of  $S_1$  offspring on parents was 62.21 percent at  $60^\circ\text{F}$ . Estimates of similar magnitude were obtained when resistance was measured by adult aphid damage. All parent-progeny and interprogeny correlations for the same and different resistance criteria were highly significant at both temperatures but were higher at  $85^\circ$  than at  $60^\circ$ . For example, the correlation between aphid survival on  $S_1$  progenies and survival of  $S_1$  progenies after infestation with aphids was -0.54 and -0.94 at  $60^\circ$  and  $85^\circ$ , respectively. The lower correlations at  $60^\circ$  appeared to result from loss of antibiosis-type resistance but retention of tolerance- or nonpreference-types of resistance by several clones at  $60^\circ$ .

7. Stem nematode resistance. In North Carolina, preliminary forage yield tests of a stem nematode-resistant synthetic from the cross C900 x Flamande backcrossed twice to Flamande indicated improved performance on both infested and noninfested soils.

8. Multiple pest resistance. Nev. Syn T, which was bred for resistance to spotted alfalfa aphid, pea aphid, stem nematode and bacterial wilt continued to perform well in irrigated areas of the Far West. Significant progress was made in combining (1) spotted aphid and pea aphid resistance with desired characteristics of Flemish alfalfas and increasing spotted aphid resistance in a susceptible winter hardy alfalfa in Nevada; and (2) resistance to spotted alfalfa aphid, pea aphid, leafhoppers, and bacterial wilt with winter hardiness in Nebraska, and resistance to spotted alfalfa aphid, pea aphid and bacterial wilt with drought tolerance in Kansas.

9. Plant pigments in alfalfa - basic color factor "C." At St. Paul, Minn., a diploid cross ( $Cc Pp Yx_1yx_1 Yx_2yx_2$ ) x ( $cc PP yx_1yx_1 yx_2yx_2$ ) segregated approximately 1:3:1:3 for purple, variegated, white and cream flowered individuals. 'C' was postulated to be a basic factor for both anthocyanin and yellow pigments in the flower. Plants with cream flowers were unexpected but assumed to have resulted either from the inability of the  $cc$  genotype to completely suppress one of the genes for xanthophyll production or from another gene producing small amounts of another yellow pigment. Investigations at the tetraploid level indicated that the 'C' gene also inhibited expression of the 'Rd' gene which controls anthocyanin in roots.

10. Self-pollination problem. Further research substantiates the urgency of developing mechanisms for assuring cross-pollination in alfalfa in order to make maximum use of heterosis. At Lincoln, Nebr., 4 clones homozygous for flower color, one white and 3 purple, and each about 30 percent self-fertile, were intercrossed with honeybees in screened cages. Flower color of the progeny from the recessive white-flowered clone indicated that only about 57 percent crossing occurred with the use of a nucleus colony and 62 to 72 percent crossing with a hive and connector.

11. Uncontrolled self-pollination reduces yield in broadcast stands. Two years of forage yield data from a Reno, Nev., experiment of a 2-clone combination admixed with varying amounts of  $S_1$  seed from each of 5 clones and seeded at different rates per acre indicated that all main effects except seeding rates were significant and that plots containing as little as 10 percent  $S_1$  seed yielded significantly less than the undiluted 2-clone combination.

12. Controlled environments for developing resistance to common leaf spot. At University Park, Pa., recurrent phenotypic selection conducted in a growth chamber made it possible to evaluate one generation of plants for resistance to Pseudopeziza medicaginis in 6 months and move from moderately susceptible to highly resistant levels in 3 generations. An average of 8,000 plants were tested in each generation. To have tested the same number of plants and the same number of generations in the field would have required 36 months. The growth chamber also gave more dependable disease infection than would be expected in field studies.



13. Interspecific hybridization. At St. Paul, Minn., attempts were made to culture several hundred embryos from a cross of  $2n = 16$  Medicago sativa x  $2n = 16$  M. pironae. Although no plants were grown to maturity, the results indicated that with the right conditions this cross could be made. An  $F_1$  cross of M. tunetana x Minn. 247 was almost free of blackstem infection in the field; whereas, nearby susceptible plants were badly infected.

14. Components of genetic variance. The components of genetic variance for seedling vigor, recovery after cutting, procumbence, resistance to downy mildew, and fall growth were estimated in two populations of double crosses in alfalfa grown at Ithaca, N. Y., assuming autotetraploid inheritance. Almost half of the estimates of genetic variance were negative. The pattern of negative estimates was very consistent; either additive and trigenic, or digenic and quadrigenic estimates were negative. The consistency of the pattern indicated that something in addition to chance was influencing the negative estimates. Evidence was found that autotetraploid epistasis or diploid inheritance with epistasis may have been partially responsible.

#### B. Diseases

1. Response of alfalfa clones to alfalfa mosaic isolates. Field and greenhouse tests of 11 alfalfa clones inoculated with 5 alfalfa mosaic isolates at St. Paul, Minn., showed that clones susceptible in the greenhouse were susceptible to the same virus isolates in the field, though not necessarily to the same extent. The clones varied in the ease with which they were infected and the virus isolates varied in their infectivity in the clones.

2. Strains of Rhizoctonia solani classified. Five groups of the alfalfa pathogen Rhizoctonia solani were distinguished within a large collection of mycelial isolates from diseased plants when correlations among morphologic, physiologic, and pathogenic traits were considered simultaneously. In the research at Raleigh, N. C., no single character separated all groups and some characters were not group specific. Pathogenicity tests on seedlings and leaves of 7 hosts showed little evidence for specialization. Tomato and wheat seedlings, wheat leaves, and emerged cabbage seedlings were more resistant than alfalfa, Korean lespedeza, lettuce, and beet to virulent isolates. R. solani groups A and B corresponded, respectively, to the imperfect stages of Pellicularia filamentosa f. microsclerotia and P. filamentosa f. sasakii. Two groups, designated C and D, were distinguished in P. filamentosa f. solani. R. solani group E corresponded to the imperfect stage of P. praticola.

3. Procedure for determining resistance to Verticillium wilt. Research in Italy revealed that scoring a cross-section of root for xylem discoloration proved to be a more reliable index to resistance than one based on wilting and stunting. Comparisons of Verticillium albo-atrum and V. dahliae showed that the former only was capable of infecting Medicago species.

4. Pathogenicity of Fusarium oxysporum medicaginis independent of potassium level. A greenhouse study conducted at University Park, Pa., showed that at low K level, plant dry weight and percentage of K in plant tops were reduced; however, disease severity did not differ at high or low levels of K. The 6 alfalfa strains evaluated differed in disease susceptibility. Only twenty percent of the plants in N.C. MSA were killed by the pathogen compared with 53 percent in Du Puits.

### C. Culture and Physiology

1. Bioassay for quantity and quality of saponin. At Logan, Utah, carrot, lettuce, cotton and several fungi were investigated as bioassayants for saponin. Of the plants tested, carrot and lettuce were the most suitable. The fungus Trichoderma sp. was a good indicator. Saponin concentrations as low as 0.02 percent reduced Trichoderma growth by 60 percent. Earlier work demonstrated quantitative differences in saponin content among varieties of alfalfa. Current research suggests qualitative differences as well. Only about 40 percent of the saponin is toxic to the Trichoderma fungus.
2. Biosynthesis of disease-induced coumestrol under study. Detached leaflets inoculated with Uromyces striatus at Raleigh, N. C., had no detectable coumestrol 3 days after inoculation, a small amount was detected at 6 days, and 7 to 104 ppm (dry wt. basis) was found 10 days after inoculation. Coumestrol was not found in healthy controls. Plants inoculated with Ascochyta imperfecta first contained coumestrol 6 to 9 days after inoculation. No coumestrol was found in autoclaved alfalfa stems on which A. imperfecta was cultured, or in mycelium of A. imperfecta grown in liquid media, confirming a host-pathogen relationship for coumestrol synthesis.

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CLOVER AND OTHER LEGUMES, CULTURE,  
BREEDING AND GENETICS, DISEASE, AND VARIETAL EVALUATION  
Crops Research Division, ARS

Problem. Emphasis within this area of research is directed mainly toward the improvement of red, white, alsike, and crimson clovers; white and yellow biennial sweetclovers; Korean and sericea lespedeza; birdsfoot trefoil; crownvetch; and blue lupines. At least 50 other legume species are either of regional or national importance or are potentially valuable for forage, provided adapted palatable varieties can be developed. One or more of these are grown on most farms or ranches either for pasture, hay, silage, soil conservation, or multiple uses. Lack of adaptation to adverse climatic conditions coupled with losses due to diseases and insects can reduce farm value of these legumes by 50%. All Trifolium species of current economic importance lack persistence and resistance to specific diseases and insects. Interspecific hybridization, possibly the key to further plant improvement in Trifoliums, has been extremely difficult to date. Sweetclover improvement objectives include low coumarin, large seed, and resistance to sweetclover aphid and weevil. Improvement in trefoil with respect to seedling vigor, resistance to root rots, broader adaptation, and greater productivity as a grazing plant on soils having poor internal drainage are important objectives. More nutritious (low tannin) and more productive varieties of sericea lespedeza are needed for eroded, acid soils of the South. Lupines, to be a reliable crop for winter grazing, must be more winter-hardy, low in alkaloids, and resistant to diseases and insects. Greater mid- and late-summer production in permanent pastures is the primary objective in the improvement of crownvetch, a long-lived, strongly creeping, rooted legume widely used for erosion control.

USDA AND COOPERATIVE PROGRAM

The Department has a long-time basic and applied research program underway involving research workers largely in cooperation with State Agricultural Experiment Stations. Research involves breeding, pathological or physiological studies on red clover at Madison, Wis.; University Park, Pa.; and Lexington, Ky.; sweetclover at Lincoln, Neb.; and Madison, Wis.; white clover at Clemson, S. C.; and Durham, N. H.; crimson clover at State College, Miss.; other perennial clovers at Ft. Collins, Colo.; other winter annual clovers at Beaumont, Texas; birdsfoot trefoil at Ithaca, N. Y.; Columbia, Mo.; Blacksburg, Va.; Stoneville, Miss.; and Tifton, Ga.; lupines and tick clover at Tifton, Ga.; lespedeza at Raleigh, N. C.; and crownvetch at Raleigh, N. C.; Columbia, Mo.; and Blacksburg, Va. In addition to the cooperative program, basic and applied research involving breeding and/or disease investigations are underway on red clover, birdsfoot trefoil, and crownvetch at Beltsville, Md. Most entomological research is in cooperation with the Entomology Research Division.

Physiological investigations on tannins and alkaloids of legumes at Experiment, Ga., were terminated June 30, 1964. Improvement of winter annual clovers at Beaumont, Texas, was terminated during 1965. Slight reductions in trefoil programs at Blacksburg, Va., and Columbia, Mo., have been made to permit limited crownvetch improvement research.

Five-year (1961-66) contracts with appropriate research departments in foreign countries, supported in part by P.L. 480 funds, involve research on: (1) Metabolism of alkaloids in legumes and their physiological role, Poland; (2) forms of nitrogen in legumes, Poland; and (3) collection and evaluation of native and introduced legumes, Brazil. In addition, a 3-year contract on virus diseases of leguminous plants in Spain was completed. A 4-year contract on polyploidy breeding in clover species in Finland was terminated.

The Federal scientific effort devoted to this research totals approximately 19.0 professional man-years, with 10.7 devoted to breeding and genetics, 2.7 to culture and physiology, 4.7 to diseases, and 0.9 to quality and varietal evaluation.

#### PROGRAM OF THE STATE EXPERIMENT STATIONS

The research program of the States relating to clover and other legumes involves the coordinated efforts of geneticists, plant breeders, plant physiologists, pathologists and agronomists, as well as scientists in other fields of endeavor. The work is both basic and applied with recognition for the need for a balanced effort.

Currently, attention is being focused on red clover, sweetclover, crimson clover, white clover, lespedeza, trefoil, and crownvetch. Other legumes show promise in a few States and exploratory research covers a broad spectrum of plants. Periodic meetings and coordination of research between states is facilitated by regional research projects such as NC-7 (Plant introduction), NC-64 (Forage quality), NE-24 (Forage nutritive evaluation), NE-28 (Forage crop breeding), NE-29 (Forage crop management), S-9 (Plant introduction), S-45 (Nutritional evaluation of forages), S-46 (Breeding forage crops), S-47 (Environment of forage crops), and W-58 (Forage crop production).

Investigations on diseases of clover, lespedeza, and trefoil are in progress at a number of the State Stations. Research on virus diseases of white clover may lead to improved stolon development in this crop. The role of light in the sporulation of two fungal pathogens of clover, Leptosphaerulina trifolii and H. australis, is being determined; and genetic materials for use in breeding studies to obtain resistance are being developed. Research on specialized inoculation techniques with root knot nematode and powdery mildew provides a new approach to obtaining resistance to these diseases in white and red clover. Multigenic resistance to these nema species affecting red clover offers much promise. The role of



root reserves in root diseases of trefoil, with special emphasis on the fungal pathogen, Leptodiscus terrestris, is being investigated. New knowledge on the mechanism of flower bud necrosis in trefoil will help to provide control of this problem. Detailed cytological studies on Heterodera trifolii and H. lespedezae are providing new knowledge on these nema cyst forms. Leadership in new research on the endotrophic fungi of red clover, such as species of Endogone, indicates pronounced beneficial effects can be obtained through the use of these fungi.

The total research program of the States involves approximately 45.8 professional man-years of which 10.0 is for culture, 23.9 for breeding and genetics, 10.5 for diseases, and 1.4 for variety evaluation.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Breeding and Genetics

1. Red clover. Synthetic varieties in first generation of synthesis were not superior to their corresponding second generation of synthesis in Wisconsin evaluations during year of establishment. Lines were screened in the greenhouse for resistance to mildew, northern anthracnose, and three virus diseases. Seven of eight leaf-mark types were determined by alleles at a single locus; the remaining type was influenced by non-allelic genes. Diploids and corresponding tetraploids differed slightly for reaction to northern anthracnose. Field evaluation of breeding materials selected for persistence was initiated in Pennsylvania; all available plant introductions were established at Beltsville for screening for morphological types and for adaptation.

In a P.L. 480 project in Finland, selection for increased fertility within each of 2 established tetraploid red clovers had little effect; such selection was more effective within each of 2 newly created tetraploid red clovers.

2. Sweetclover. Cooperative research in Nebraska with the Entomology Research Division screened 52 additional entries for sweetclover weevil resistance. High levels of resistance were found in Melilotus infesta and M. sulcata; some plants of M. polonica were intermediate in resistance. Aphid resistance was present in 20 to 25 percent of 15,000 seedlings from various breeding materials; more than 90 percent of self progeny of selected resistant plants were resistant to the aphid. Biochemical research showed that increases in temperature, light intensity, and photoperiod to which plants were exposed were generally followed by increases in amounts of o-hydroxycinnamic acid in leaves, stems, and roots of plants. The sweetclover weevil resistance factor apparently moved across the graft union of M. infesta and M. officinalis 6 weeks after grafting. The B allele appears without dominance effect for B-glucosidase activity. Phenylalanine deaminase activity was detected in both Cu Cu and cu cu genotypes; attempts to demonstrate in vitro



conversion of trans-cinnamic acid to o-coumaryl glucoside and the deamination of o-tyrosine to o-coumaric acid were unsuccessful. Embryos were found only in crosses of M. infesta with M. sulcata and M. italica. Additional evidence was obtained indicating that little contamination of low coumarin varieties with high coumarin germ plasm occurs under seed certification procedures. Approximately 50 percent cross-pollination occurred among adjacent plants of high and low coumarin varieties. In Wisconsin, studies with F<sub>1</sub>, F<sub>2</sub>, and F<sub>3</sub> generations of crosses between low coumarin M. dentata and high coumarin M. taurica support the hypothesis that B-glucosidase and coumarin contents are each determined by single genes.

3. Crimson clover. In Mississippi inbred lines produced polycrosses superior to the parental variety from which inbreds were derived for seasonal production of forage, disease resistance, and seed production. Genetic studies on qualitative characteristics were continued.

4. White clover. Forage production and stands of plants with adventitious roots only or complete root systems (primary plus adventitious) were similar in South Carolina growth chamber and field studies. Clones tolerant to root-knot nematode were intercrossed to produce new populations for evaluation. Attempts to cross a recently acquired species, T. occidentale, with white clover are in progress.

5. Winter annual clovers. Selection was continued at Beaumont, Texas, for hard seed and winterhardiness in berseem clovers; progeny of 121 plants constitute the population for future work.

6. Perennial clovers. In Colorado a self-compatibility factor inherited as an allele to the multiple S series was found in diploid alsike clover. Self-compatibility of a tetraploid alsike clone was influenced by temperature: Higher temperatures increased self-compatibility; the self-compatibility reaction was reversible with appropriate temperature changes. The site for change in the compatibility reaction was found to be in the pistil.

7. Birdsfoot trefoil. In cooperative studies at Ithaca, N. Y., HCN presence has been successfully used as a dominant genetic marker in studies of methods of clone synthesis of importance in the development of new varieties. Seed and seedlings produced by selfing are smaller than those produced by outcrossing. In this same program six inter-specific crosses involving L. corniculatus as the male parent have been backcrossed to L. corniculatus three times in an effort to produce cytoplasmic male sterile types. Maximum male sterility after two backcrosses was 70 to 80%. Greater longevity and root-rot resistance is being obtained in populations of trefoil in the cooperative programs at Columbia, Mo., Blacksburg, Va., and at Beltsville, Md., through recurrent phenotypic selection. A total of 17 new synthetics developed in these programs are being evaluated in field tests. An effort to develop

a variety of trefoil adapted to more southern locations is being made. Selected nonhardy plants introduced from South America have been hybridized with good domestic clones, and F<sub>1</sub> and F<sub>2</sub> progenies are being grown at a number of southern locations.

8. Lupines. Excellent progress is being made in the development of more winter-hardy, disease-resistant forage varieties of blue lupine in the cooperative program at Tifton, Ga. Concurrent greenhouse disease inoculations and laboratory studies have been conducted to identify elite homozygous lines with respect to anthracnose and gray-leaf spot resistance and soft seededness. Some new winterhardy strains will be available for testing in the Fall of 1965. No linkage was found between hard seededness and alkaloid content. The new variety, Rancher (20-206), will be released for seed production in the fall of 1965. Two hundred pounds of seed of homozygous sweet yellow lupine were produced by using systemic insecticides to control vectors of the virus of this species.

9. Crownvetch. This long-lived, creeping rooted, perennial legume shows excellent potential as a forage legume for permanent pastures. Limited breeding programs have recently been initiated in North Carolina, Missouri, and Virginia. Crownvetch has been found resistant to root-knot nematodes at Raleigh, N. C. Progenies of 192 plants are being evaluated for palatability and forage production at Beltsville. Productive, strongly creeping rooted clones are being selected for further evaluation in these programs.

10. Sericea lespedeza. Selection following two intercrossing cycles of low versus high tannin plants is underway at Raleigh, N. C., to secure superior productive and nutritious lines of Sericea lespedeza. A number of low tannin (palatable) lines are under test. Less than 10% natural crossing occurred in Sericea in the field in North Carolina. This level of crossing raises a serious question concerning the importance of heterosis in developing new varieties of this species.

11. Desmodium sp. One hundred thirty seed accessions representing 16 Desmodium species have been evaluated in the cooperative program at Tifton, Ga., in an effort to find a productive, perennial forage legume for southern pastures. The majority of accessions tested have little if any value for the area due either to winter-killing, low forage value, or susceptibility to diseases. However, two species, D. uncinatum and D. intortum, appear promising. All accessions of the former were winter-hardy and quite productive. Further evaluations are being made of these species in association with coastal bermudagrass. Other perennial legumes including Lotononis bainesii, Phaseolus atropurpureus, and Glycine javanica which have shown promise in other areas are being studied.

12. Korean lespedeza. A new strain of Korean lespedeza, NC-24, developed in cooperation with North Carolina was clearly superior to other



varieties in 1964 tests. NC-24 has a high degree of resistance to tar spot, root-knot nematodes, and produces high forage and seed yields. Foundation seed is being produced for release of the variety in 1966.

13. Big trefoil. In absence of diseases this species, Lotus pedunculatus, has appeared promising as a perennial pasture legume in the Southern States. Stands of big trefoil in the South are either killed by Rhizoctonia or other pathogens or reduced to the point where they are no longer productive. Cooperative studies at Tifton, Ga., Stoneville, Miss., and Blacksburg, Va., involving selection and recurrent selection to secure disease resistant plants have not been successful to date. The limited programs with this species are to be terminated in 1965.

#### B. Diseases

1. Incidence of internal breakdown of red clover correlated with crown diameter. Previous experiments in Pennsylvania indicated no direct effect of soil fertility or pathogenic agent on incidence of internal breakdown; recent pooling of all previous experimental data indicated a positive correlation between incidence of internal breakdown and crown diameter, independent of age of plants. New experiments with daylength, temperature, clipping severity, and plant density variables gave highest incidence of internal breakdown for treatments most favorable for plant growth. Effects of thermal neutron irradiation of seed and seedlings on incidence of internal breakdown were inconclusive but suggested increase in incidence of internal breakdown.

2. Cultural characteristics of 2 fungi determined. In cooperative studies at South Carolina, cultures of Leptosphaerulina briosiana predisposed at 20°C sporulated less than cultures predisposed at 22°C. Sporulation increased on exposure to light; blue, green, and white light induced more sporulation than red light. Ascospores were ejected and germinated under all conditions. Conidia of Pleospora herbarum were produced most abundantly on V-8 juice agar under green light.

3. Virus found in birdsfoot trefoil. A disease infection, tentatively identified as the tobacco ring spot virus was found in at least 2 of 21 field grown plants of birdsfoot trefoil at Beltsville, Md. Except for a general unthriftness and a mild chlorosis, plants were symptomless. The synergistic effect of virus infection and root-rotting fungi in trefoil is being studied.

4. A new root-rotting fungus of birdsfoot trefoil. A new sclerotial fungus described previously is believed to be a new species of Leptodiscus. Sparse sporulation occurred in culture during the summer. Single spore cultures were identical to parent cultures on differential medium. Efforts will be made to reisolate this organism in the field.



5. Crownvetch resistant to root-knot nematode. Varieties of crownvetch were found to have strong resistance to the root-knot nematode species, Meloidogyne incognita, M. javanica, and M. arenaria, in inoculation studies in the cooperative program at Raleigh, N. C. Crownvetch was moderately susceptible to M. hapla; however, none of the nematode species caused marked root damage or above ground symptoms. Reactions appeared quite uniform within and between varieties.
6. Polyphenols reduce activity of enzymes produced by Rhizoctonia solani. The enzymes, Pectin transeliminase and polygalacturonase, produced by R. solani in culture were partially inhibited by a high molecular weight polyphenol from Sericea lespedeza in studies at Raleigh, N. C. Inhibition was greater when the inhibitor was mixed with the enzyme before the enzyme was mixed with the substrate. Monophenols, such as catechol, did not inhibit pectic enzymes; however, monophenols oxidized by shaking in air were inhibitory.
7. Two types of resistance to tar spot in annual lespedeza. Two different anatomical manifestations of resistance to tar spot were found in studies at Raleigh, N. C. In one type the outer wall of the epidermal cell beneath the fungus appressorium becomes so thickened that in most cases the penetration pegs are unable to pass through the thickened wall. Infection cannot occur without penetration of the cell wall. This type of resistance was typical of the resistant variety Kobe. The frequency of wall thickening in 4 varieties of Korean was correlated with their apparent resistance in the field. In the second type of resistance, invaded areas of the leaves collapsed and died two weeks after inoculation and before the fungus could develop further or fructify in the dead areas. The reaction appeared to be due to hypersensitivity.
8. A source of resistance to brownspot in lupines. Yellow lupine, Lupinus luteus, are very susceptible to brown spot caused by Pleiochaeta setosa. In two consecutive years L. hispanicus and L. rothmaleri grown in association with severely diseased L. luteus were free of brown spot. Interspecific hybridization of the two resistant species with L. luteus is being explored in the program at Tifton, Ga.
9. New diseases found in vetch. Two previously unrecorded diseases were observed on narrow-leaved vetch at Stoneville, Miss., in 1964. Phytophthora root and crown rot caused by P. erythroseptica attacked woolypod vetch, white lupine, Romack winter pea, Alaska garden pea, and narrow-leaved vetch when planted on infected soil. A species of Physoderma caused hyperthrophy of the basal buds and galling of stems and leaves of narrow-leaved vetch. The symptoms on vetch resembled in some respects those of crown wart disease of alfalfa. This is the first report of these diseases occurring on vetch in the United States.

10. Legume virus inclusion bodies characterized. Research on a P.L. 480 project in Spain showed that inclusion bodies of amorphous type were formed of virus particles and extraneous amorphous material. The percentage of amorphous material in amorphous type inclusion bodies varies with kind of virus and age. Several kinds of amorphous and crystalline inclusion bodies were characterized with respect to shape and composition.

#### C. Quality and varietal evaluation

1. Clovers. Synthetic varieties formulated in Wisconsin were superior to existing varieties for forage yields in the second harvest year. A re-seeding crimson clover variety, with agronomic characteristics similar to Frontier, is available for release from Mississippi. A white clover synthetic from South Carolina excelled in both South Carolina and regional tests. Uniform tests of sweetclovers were conducted at 10 locations in the United States and Canada. Rustler was highest yielding of berseem varieties tested at Beaumont, Texas.

#### D. Culture and physiology

1. Red clover. A technique for measuring foam production as related to bloat was refined at Kentucky, allowing detection among clones in foaming potential. Differences were not detected among 8 varieties for reaction to various concentrations of 5 herbicides. Sensitivity of detached leaves to a heat treatment was lessened by a pretreatment of brief exposure to a relatively high temperature; leaves of most persistent clones were most subject to heat injury.

2. White clover. Carbohydrate, free amino acid, and total protein changes were determined for seed germination and early seedling stages of growth in South Carolina studies. Morphological, protein, carbohydrate and nucleic acid changes with time, at controlled temperatures ranging from 16 to 40°C, were determined for seedling shoots. Necrosis of surface tissues was characteristically found in bud regions of nodes of a clone known to be relatively inactive in axillary bud development.

3. Crimson clover. Inbred lines that germinate only under low temperatures were identified in Mississippi. Most such lines were late in maturity.

4. Toxic nonprotein amino acids in Lathyrus sp. Biochemical studies conducted in Poland, revealed that 9 of 17 Lathyrus species contain the toxic nonprotein amino acids responsible for "Lathyrism" in man and animals. While only the seed of some species contained toxic substances, all plant parts of L. sylvestris and L. latifolius were toxic. The above species grow well in northern USA and have been used for erosion control. The rather widespread occurrence of toxic amino acids in Lathyrus indicates that it would be extremely difficult to develop a palatable forage variety of either L. sylvestris or L. latifolius.



5. Alkaloid content of legumes affects nitrogen content. High alkaloid containing lupine plants yield more and contain more nitrogen than the low alkaloid or fodder varieties. As to form of nitrogen, the fodder varieties contain significantly less arginine than high alkaloid lines.

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FORAGE GRASS AND TURF CULTURE,  
BREEDING, DISEASES, AND VARIETAL EVALUATION  
Crops Research Division, ARS

Problem: Some 90 introduced and native grasses are of agricultural importance in the United States for forage, turf, and soil conservation purposes. Of these, only 25 are included in active research programs. Up to 50 percent of new seedlings are failures or near failures; and this staggering loss could be reduced materially with varieties possessing improved seedling vigor, seedling drought tolerance, and disease resistance. Diseases reduce the yield and quality of forage and seed and contribute to the loss of established plantings grown for either forage or turf purposes. More research is needed to overcome these seeding, establishment, and disease hazards. In most grasses, progress in developing improved varieties depends on the accumulation of basic information in the fields of cytology, genetics, pathology, and physiology. Problems associated with management and maintenance of turfgrass areas are poorly understood. More than three billion dollars are spent annually in establishing and maintaining turf. Improved management practices and improved grass varieties are urgently needed to meet the different requirements of use, climate, and soil that characterize the major turfgrass regions of the United States.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving agronomists, geneticists and plant pathologists engaged in both basic and applied research. Forage grass and turf research is underway at Tucson, Ariz.; Tifton, Ga.; Manhattan, Kans.; Lexington, Ky.; Beltsville, Md.; State College and Stoneville, Miss.; Lincoln, Nebr.; Mandan, N. Dak.; Woodward, Okla.; University Park, Pa.; College Station, Tex.; Logan, Utah; and Madison, Wis. All work is conducted in cooperation with the respective State Agricultural Experiment Station. Research on range grass breeding at Laramie, Wyoming, has been terminated. A new position was established at Tifton, Georgia, to develop insect-resistant bermudagrass varieties.

Four PL 480 projects have been negotiated in India; namely, Cataloguing and classifying genetic stocks of Pennisetum spp.; The establishment of linkage groups in Pennisetum typhoides; Pollen storage and serological classification; and Genetic evaluation of grain and fodder quality in millet. A PL 480 project in Rio Grande do Sul, Brazil, involves ecological and cytological studies and genetic improvement of forage grasses and legumes.

The Federal scientific effort devoted to research in this area totals 17.9 professional man-years. Of this number 13.4 are devoted to breeding and genetics, 1.2 to diseases, .9 to quality and varietal evaluation, and 2.4 to cultural investigations.

## PROGRAM OF STATE EXPERIMENT STATIONS

The State Agricultural Experiment Stations are engaged in many research activities relating to forage breeding and evaluation. The intensity of this work and the division between basic and applied research are variable. Generally, those States with the largest programs are more inclined to support basic efforts; whereas, other States are limited to routine testing or applied research. As the program becomes more refined and the objectives more readily recognized, the role of the plant breeders has increased, going beyond selection to actual breeding and hybridization. Crosses and new combinations that were considered impossible a few years ago are now being routinely made.

Attention is being focused on evaluation techniques relating more directly to animal performance. The standard chemical and biochemical measurements are valuable but do not serve as adequate guidelines for animal response. At the same time, research is continuing on disease resistance and adaptation to specific soils and climates. Research on cultural practices and management techniques is also a part of this forage program.

Turfgrass research is expanding with new projects being initiated in more States. Interest in this area is growing rapidly and will require the combined efforts of agronomists, geneticists, pathologists, and other members as scientific teams.

Research between States is effectively coordinated through regional research projects such as NC-7 (Plant introduction), NC-64 (Forage quality), NE-24 (Forage nutritive evaluation), NE-28 (Forage crop breeding), NE-29 (Forage crop management), S-9 (Plant introduction), S-45 (Nutritional evaluation of forages), S-46 (Breeding forage crops), S-47 (Environment of forage crops), and W-58 (Forage crop production).

Scientists at the State Stations are involved in research on a number of disease problems in the culture of grasses. Three research groups are concerned with studies on the role of fungi in the production of toxins in fescue and bermuda. Research is in progress on a number of diseases of orchardgrass. Recent findings in studies on rathayi disease on orchardgrass has provided useful knowledge on the relation of this bacterial pathogen to forms causing serious diseases of grain. Such work is fundamental to control of these destructive diseases. Studies on diseases of sudangrass and resistance to Sclerotinia blight in bahiagrass are being pursued through effective research at two State Stations. Germplasm to provide efficient resistance to crown rust and Helminthosporium blight of ryegrass, Rhizoctonia blight in fescue, leaf and stem blight in bermuda, and rust in bluegrass is being isolated and evaluated in several research projects.

The total research program of the States involves approximately 145.3 professional man-years of which 60.7 is for culture, 59.7 for breeding and genetics, 8.6 for diseases, and 16.3 for variety evaluation.



PROGRESS -- USDA AND COOPERATIVE PROGRAMS

A. Breeding and Genetics

1. Regional Kentucky bluegrass collections characterized. Collections of Kentucky bluegrass clones from three major sites in both Alabama and Maryland were characterized for climatic, edaphic, and management variables in a clonal nursery and two progeny tests at Beltsville, Md. Chi-square values for associations between Alabama and Maryland collections were highly significant for leaf rust, leaf spot, recovery from leaf spot damage and apomixis. Snow mold injury was not significantly associated with the two collection regions. Alabama collections included a higher proportion of apomictic clones and clones resistant to leaf rust. In Maryland and the mid-Atlantic region leaf spot tolerance makes a substantial contribution to the persistence and performance of Kentucky bluegrass varieties. The greater frequency and higher levels of leaf spot-tolerant isolates found in Maryland would increase the value of local collections over those made in Alabama, outside the accepted range of adaptation for Kentucky bluegrass.

2. Promising range plants developed by crossing sexual x apomictic buffelgrass. Basic research at College Station, Texas, resulted in the development of a successful breeding program in a predominantly apomictic species--buffelgrass. It was shown that apomixis can be manipulated to permit the mass production of true breeding F<sub>1</sub> hybrids and new strains. Hybrids between a sexual plant and two apomictic lines segregated in a 1:1 ratio for sexual and apomictic plants. Progeny from some crosses deviate from this ratio. Available data suggest that obligate apomixis is controlled by no more than two genes. Field evaluation of 25 new apomictic strains in 1964 showed that some were superior to the best commercial variety in seed and forage yield.

3. Tifdwarf bermudagrass released. Tifdwarf bermudagrass developed at Tifton, Ga., was released, in cooperation with the Georgia Agricultural Experiment Stations, for use on golf greens and other areas where high quality turf is needed. Available evidence suggests that Tifdwarf is a vegetative mutant that occurred in the Tifgreen variety before the first stocks of Tifgreen were distributed. Tifdwarf is similar to Tifgreen in many respects, but is characterized by shorter leaves, lower growth habit, and a darker green color. Tifdwarf spreads faster than Tifgreen and requires much less top dressing to maintain a good-appearing, smooth, putting surface. Tifdwarf maintained at 3/16- and 1/4-inch heights produced only about half as many clippings by weight as Tifgreen. These findings suggest that the amount of mowing and hence, labor costs could be reduced by using Tifdwarf.

4. Relationship of coleoptile length to emergence in intermediate wheatgrass. Better stand establishment could be expected if varieties

with improved emergence potential could be developed for range reseeding. These varieties would emerge better from deeper seeding often required to reach moisture. Investigations with intermediate wheatgrass at Laramie, Wyo., suggest that good emergence is closely related to seed weight and coleoptile length. Weight of 200 seed when compared with coleoptile length and emergence from a 3-inch planting depth gave correlation coefficients of .805 and .80, respectively. Analyses of diallel crosses indicated a strong maternal influence on the inheritance of coleoptile length. However, long coleoptile females produced progeny with longer coleoptiles when crossed with long coleoptile males than with short coleoptile males. This suggests that improvement in coleoptile length, emergence potential, or seed size could be accomplished through simple maternal line selection with open-pollinated progenies. The effectiveness of selection based on seed size alone may be reduced by environmental effects.

5. Inbreeding depression in crested wheatgrass. Although inbreeding depression has not been adequately evaluated in any form of crested wheatgrass, available data indicates that the effects of inbreeding are at least as severe in tetraploid forms as in diploids. This has been explained by the assumption that the species is a segmental allopolyploid, and therefore should inbreed much the same as a diploid. However, cytogenetic information supports the viewpoint that crested wheatgrass is a strict autopolyploid. Data accumulated at Logan, Utah, may help to explain this anomalous breeding behavior. The effect of inbreeding on vegetative vigor and fertility was evaluated in diploid, tetraploid, and hexaploid  $S_1$  progenies. Forage yields of  $S_1$  progenies averaged 64.1, 49.6, and 32.6% of comparable open-pollinated yields of diploid, tetraploid, and hexaploid populations, respectively. Open-pollinated seed set on these same  $S_1$  populations averaged 47.8, 57.9, and 31.0% of their open-pollinated counterparts. It is suggested that severe inbreeding depression in tetraploid and hexaploid populations may be attributed to chromosome aberrations that accumulate in species buffered by autopolyploidy.

6. Chance-hybrid varieties for range reseeding. Diverse genotypes of little bluestem, big bluestem, sand bluestem and switchgrass have been used to develop synthetic varieties at Lincoln, Nebr. The production of chance-hybrids is being investigated at present, as a method for increasing seedling vigor, persistence, and productiveness in range grass varieties. In this procedure good combining ecotypes that are similar in maturity are maintained in separate isolation blocks. In little bluestem, progeny with about 40-50 percent hybrid plants were obtained from isolation blocks in which two similar maturing ecotypes were planted in alternate rows.

7. Short-day sensitive pearl millet for improved quality. Photo-periodism was exhibited by 40 out of 290 pearl millet accessions received from Nigeria and Upper Volta. In tests conducted at Tifton, Ga., these 40 short-day sensitive lines reached anthesis in November whether planted in May or August. When planted in December or January in the greenhouse, they flower in about the same number of days as day-neutral lines. The other 250 introductions, like most pearl millet lines in the Tifton collection, were day-neutral and flower in 77 or 87 days. Hybrids between short-day sensitive lines and day-neutral Tift 23A showed heterosis, were intermediate in maturity, and flowered in late September. Late-maturing millets should be superior to early millets in leafiness, seasonal distribution of forage produced, and ease of management. Such millets, planted late in the season, should mature seed at heights of 3 to 4 feet and be well suited to commercial seed production by combine harvesting.

8. Pollen storage facilitates hybridization in pearl millet. The discovery at Tifton, Ga., of cytoplasmic male-sterility in pearl millet has led to increased hybridization studies in this species, with the resulting need for additional information on pollination techniques. Studies were conducted at Tifton, Ga., on the effects of pollen storage and hour of pollination on seed set. It was found that pearl millet pollen may be stored 24 hours at 80°F and up to 4 days at 40°F with less than 50% loss in viability. Very little pollen survived storage at 80°F for two days. Pollen stored at 40°F for three weeks gave only 8% as good seed set as fresh pollen, and all pollen stored at this temperature for four weeks was dead. Fresh pollen applied at 9:30 A.M., 1:30 P.M., and 4:30 P.M. to male-sterile lines gave average seed set values of .67, .38 and .74 grams of seed per inch of head, respectively.

9. Magnolia ryegrass for the southeastern region. Magnolia ryegrass, a blend of two rust-resistant strains, will be released in cooperation with the Mississippi Agricultural Experiment Station in 1965. The two strains, Stoneville 3 and State College 7, were selected for improved resistance to crown rust and leaf spot at Stoneville and State College, Miss., respectively. The two parental strains will be maintained under isolation and the variety increased on a limited generation basis. Magnolia has a higher percentage of rust-resistant plants than do introductions from Uruguay, Gulf, or common ryegrass. It has produced good yields of forage and seed in many regional tests. At State College, Miss., two-year average yields of dry matter in tons per acre were as follows: Magnolia - 2.28, Stoneville 3 - 2.17, State College 7 - 2.15, Gulf - 1.98 and common - 1.78. The variety has performed well under grazing and can be used for overseeding warm-season turfgrasses.



10. Close relationship exists among ryegrass species. Interspecific ryegrass hybrids were obtained at University Park, Pa., by pollinating unemasculated spikes of self-incompatible plants of Lolium perenne, L. multiflorum and L. rigidum with pollen from several Lolium species. Caryopses developed from 13 interspecific combinations but failed to germinate for L. perenne x L. persicum and L. perenne x L. strictum. Fertility in the  $F_1$  and  $F_2$  generations measured less than 42% seed set. Fragment chromosomes in addition to the  $2n=14$  chromosome complement were observed at meiosis in L. remotum, L. strictum and several interspecific hybrids. Chromosome associations of 5 to 7 bivalents in most interspecific hybrids suggests that Lolium species have a common genome that is not greatly differentiated. The relatively low fertility of several hybrids indicated a restricted gene flow under experimental conditions.

11. Meiotic stability -- an important factor in breeding smooth brome. Interrelations among reproductive stages in smooth brome were investigated at Madison, Wis. Two distinct mechanisms were identified at microsporogenesis; namely, meiotic disturbances attributed to structural differences among and between chromosomes and disturbances related to metabolic errors. Metabolic errors were expressed in the form of pycnotic accumulations during prophase, stickiness at metaphase, coenocyte formations, chromosome complement fractionation and nonsynchrony of divisions. Structural differences were not related to stainable pollen or to self- and cross-pollination seed set, while metabolic errors were related to one another and to pollen abortion. It was postulated that much potential diversity in smooth brome is not expressed because of the lethal system that results from metabolic errors. Successful viable recombinations represent a relatively small portion of the available genetic diversity. These data support the view that rigid selection for self-fertility coupled with other pressures, such as disease resistance, should result in genetic recombination and advancement.

12. Genome relationships within the wheatgrasses (Agropyrons) clarified. Genome analysis of hybrids from Agropyron repens x A. riparium, A. repens x A. spicatum, and A. spicatum x A. riparium was instrumental in establishing genome relations in studies conducted at Logan, Utah. Chromosome pairing in 35 chromosome hybrids of A. repens ( $2n=42$ ) x A. riparium ( $2n=28$ ) indicated that these species share a genome in common; and this genome is responsible for the rhizomatous habit in both species. Chromosome pairing in triploid and tetraploid hybrids of A. riparium ( $2n=28$ ) x A. spicatum ( $2n=14$  and  $28$ ) showed that one of the two A. riparium genomes is closely homologous with the A. spicatum genome. Chromosome pairing in pentaploid hybrids of A. repens ( $2n=42$ ) x A. spicatum ( $2n=28$ ) indicated that these species share no common genome. If the genome formula of tetraploid A. spicatum is written as SSSS, A. riparium would be represented as  $S_1S_1R_1R_1$  and A. repens as  $R_2R_2X_1X_1X_2X_2$ . The "R" genome shared by A. riparium and A. repens probably originated from a rhizomatous diploid species. Diploid A. spicatum is the source of the "S" genome which has been identified in many Agropyron, Elymus and Sitanion species. The source and distribution of the "X" genomes of A. repens have not been established.

13. Breeding warm-season grasses for southern Brazil. Considerable variation has been found in grasses hitherto reported as obligate apomicts. Available seed stocks of Paspalum accessions from Brazil have been brought to College Station, Texas, for cytogenetic analyses.
14. Pollen storage and serological classification. Attempts to develop a suitable medium for evaluating pollen viability were successful in India. Calcium in the presence of  $MgSO_4$  and  $KNO_3$  produced good results when added to the basal medium consisting of sucrose (25%) and boric acid (0.01%). The calcium-supplemented liquid medium enhanced pollen germination and normal pollen tube growth.
15. Linkage studies in pearl millet. Material has been assembled in India and crosses made to initiate these studies.
16. Genetic evaluation of grain and fodder quality in millet. Plantings were established in India and preliminary data collected to determine variation in grain and fodder quality.

#### B. Diseases.

1. Inoculation of ryegrass with southern blight. Inoculation tests conducted at Stoneville, Miss., show that the southern blight fungus (Sclerotium rolfsii) is responsible for the loss of annual ryegrass plants in some fields. Agar-grown inoculum caused little or no preemergence killing but caused seedling blight on annual ryegrass and winter oats. Dried-grain inoculum caused both preemergence killing and seedling blight. An isolate of the fungus from birdsfoot trefoil was pathogenic in a test using dried-grain inoculum. However, no infection was obtained when the soil mixture was steam sterilized before adding the inoculum. The method of inoculation used did not reproduce the exact symptoms observed in the field. The studies were successful, however, in demonstrating that losses in some fields could be attributed to southern blight, and in showing preferred inoculation techniques for screening work.
2. A rust on zoysia new to North America. A rust, identified as Puccinia zoysiae Diet., was observed in 1964 in zoysia lawns in Wheaton, Md., and almost simultaneously in Kansas City, Mo. The rust, not previously reported in North America, may pose a new problem for homeowners with zoysia grass lawns. To date, the most widely grown variety, Meyer, has been relatively free of serious pathogens. Information is not yet available on the overwintering mechanism as the Asiatic alternate host, Paederia chinensis, has not been introduced, and the aecial stage of the rust has not been reported on P. foetida, introduced in 1936.
3. Cross infectivity and alkaloid production of Claviceps isolates. At Lexington, Ky., no consistent evidence of host-pathogen specificity was obtained among conidia from isolates of Claviceps purpurea from rye, tall fescue, annual ryegrass, tall fescue-ryegrass hybrids, and quackgrass.

Alkaloids were detected in 85% of the sclerotia collected from different host plants grown over a range of environmental conditions.

4. Billbug injury in zoysia turf. Meyer zoysia was considered to be relatively resistant to insect attack when released from Beltsville, Md., in 1951. However, in recent years the hunting billbug, Sphenophorus venatis vestita (Chttm.) has become a serious problem in zoysia turf. Injury from billbugs was reported from Florida in 1956 and first observed in the mid-Atlantic region in 1959. The organic phosphate insecticide, diazinon, gave good control of billbug adults and larvae in preliminary studies at Beltsville, Md., where damage was particularly severe in 1964. It was difficult to obtain complete control with a single application of the insecticide so that a second application in 6 to 10 weeks is often necessary. (Coop ENT)

#### C. Quality and Varietal Evaluation.

1. Kentucky bluegrass selections differ in reaction to sod webworms. In 1964 an extremely severe infestation of sod webworm at Lexington, Ky., provided an opportunity for comparing injury among experimental varieties of Kentucky bluegrass. Counts of three web worm species and damage index scores were obtained from replicated, solid-seeded plots maintained at two clipping intensities simulating turf and pasture management. Differences among varieties were significant with respect to both sod webworm larvae and pupae and damage index scores. In turf plots the average number of webworms varied from 3.25 to 7.25 per entry while the range in damage index was 1.63 to 4.75 (a score of 1 represents no damage and 5 severe damage). The correlation between infestation counts and damage index was not significant. Since the nature of the differential reaction among varieties is not known, the nonsignificant correlation coefficient could be explained on the basis of tolerance. The difference among varieties offers promise of developing varieties with resistance to sod webworm.

2. Grass variety performance not affected by seeding rate. At Mandan, N. Dak., Nordan crested wheatgrass and Vinall Russian wildrye were seeded in replicated 3-foot rows at rates of 3, 6, and 9 pounds per acre. In 1964, there were no differences among seeding rates significant at the 5% level in culm production, forage yield, seed yield, or seed size of either grass. Ranges across seeding rates for all 4 characters of both grasses were less than 5% of their respective means. Twelve-replicate tests established in 1964, using identical seeding rates, included Fairway wheatgrass and Mandan 759 pubescent wheatgrass as well as Nordan and Vinall. Initial stand densities of all 4 grasses were reflected by the seeding rates--each increase in seeding rate gave an increase in seedling emergence significant at the 1% level. For 3 of the grasses, however, there were highly significant decreases in percentage emergence from the lowest seeding rate to the highest. In Mandan 759, emergence percentages were essentially the same for all seeding rates.



#### D. Cultural Investigations

1. Kentucky bluegrass and red fescue tolerate high levels of phosphorus. The influence of above optimum levels of phosphorus on the growth and development of Kentucky bluegrass and red fescue turfgrasses was investigated at Beltsville, Md. Levels of phosphorus from 0 to 1527 pounds per acre were compared at two levels each of nitrogen and potassium, at two pH levels and on two soils. Accumulation of P in leaves of Merion exceeded that in common bluegrass. Top growth of Merion was significantly greater and root weights significantly less than common bluegrass. Results indicated that Kentucky bluegrass and red fescue can tolerate high levels of P and that upper levels of tolerance were not attained in these studies.

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PASTURE AND RANGE SEEDING,  
ESTABLISHMENT, MANAGEMENT, AND EVALUATION  
Crops Research Division, ARS

Problem: Grazing lands of the United States occupy almost one billion acres as compared to 350 million acres of all harvested crops. Presently more than half of all the nutrients consumed by domestic livestock come from pastures and ranges. With increasing population and increased demand for livestock products, the proportion of nutrients from pastures and ranges is expected to increase. Grazing lands represent a highly diverse part of our land resource and, because of their diversity, are a tremendous challenge to research. More precise information is required for many situations; information about what plants or mixtures meet the requirements for yield, nutritional value, and resistance to heat, cold, drought, and pests. Some of the major categories needing research attention are concerned with seeding and establishment, and include basic and applied physiological and ecological studies on the response of pasture and range species, biochemical constituents, and morphological development of the plant under grazing. In the years ahead, as humans compete more intensively with livestock for present feed grains, we must develop our grassland resources if this country expects to maintain high quality diets with the levels of meat and milk that are considered essential.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving agronomists, plant physiologists, range scientists, and chemists engaged in basic and applied research on the management and improvement of grazing lands, native meadows, and improved pastures. All work is cooperative with the respective States, with the U. S. Forest Service, and Bureau of Land Management, in areas where grazing is integrated with lands under their control. Research is in progress at Flagstaff, Ariz.; Berkeley, Calif.; Ft. Collins, Colo.; Gainesville, Fla.; Tifton, Ga.; Twin Falls, Idaho; Lafayette, Ind.; Beltsville, Md.; St. Paul, Minn.; Columbia, Mo.; Bozeman, Mont.; Miles City, Mont.; Reno, Nev.; Las Cruces, N. Mex.; Ithaca, N. Y.; Raleigh, N. C.; Mandan, N. Dak.; Woodward, Okla.; Burns, Oreg.; University Park, Pa.; Beaumont, Texas; Logan, Utah; and Pullman, Wash. Research at Bozeman, Mont., and Mandan, N. Dak., is conducted cooperatively with Soil and Water Conservation Research Division and at Miles City, Mont., with Animal Husbandry Research Division.

Of three P.L. 480 projects two are in Israel. One concerns developmental physiology of perennial pasture grasses. The other investigates establishment and maintenance of seeded dryland range under semiarid conditions. The third, in India, concerns basic studies on polyfructosan biosynthesis in fodder crops.



The Federal scientific effort devoted to research in this area totals 33.2 professional man-years. Of this number 6.0 is devoted to seeding and establishment, 0.2 to processing, 16.5 to management, 10.5 to quality and varietal evaluation.

#### PROGRAM OF STATE EXPERIMENT STATIONS

The program of the States relating to range and pasture seeding, evaluation, and management encompasses a wide variety of basic and applied research activities. In recent years emphasis has been directed toward more basic studies involving the understanding of principles, although surveys and other forms of applied research continue to be important. Progress also requires the combined efforts of agronomists, plant physiologists, climatologists, biochemists, and range ecologists.

Vegetation re-establishment studies on the better range and pasture lands is an important part of the research program. Species evaluation, seeding techniques, and cultural practices are studied in an overall attempt to increase the productivity of these area.

Scientists are brought together through the mechanism of regional research projects. Those regional projects which are concerned directly or indirectly with ranges and pastures include: NC-7 (Plant introduction), NC-26 (Weather information), NC-64 (Forage quality), NE-35 (Climatology), S-9 (Plant introduction), S-45 (Nutritional evaluation of forages), S-47 (Environment of forage crops), W-16 (Economics of rangelands), W-25 (Improvement of rangelands), W-34 (Range livestock nutrition), W-48 (Weather environment), and W-58 (Forage crop production).

The total research program of the States involves approximately 67.3 professional man-years of which 1.4 is for seeding, 7.2 for establishment, 35.8 for management, and 22.9 for evaluation.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Seeding and Establishment

1. Little leafage required for rapid growth of newly established plants. At Columbia, Mo., vegetatively multiplied Ky-31 tall fescue plants in the greenhouse were defoliated at 10-day intervals to a stubble height of 1-, 2-1/2-, or 4 inches, leaving 0, 10%, 20%, and 30% intact tillers. Complete defoliation to a 1-inch stubble resulted in death of the plant; however, when 10% or more intact tillers remained, tillering increased an average of 71%. Total regrowth yields and average daily growth rates were benefited by increasing the stubble height. Also, growth rate was accelerated as the number of intact tillers was increased up to 20%. Apparently 10% or more intact tillers provide additional photosynthetic surface for the production of metabolic products, or the storage of food reserves necessary for plant survival and growth under conditions of defoliation stress.

2. Birdsfoot trefoil gives highest yields on mountain pastures. In North Carolina, mountain pastures overseeded with birdsfoot trefoil was the outstanding treatment in terms of average daily gain and beef per acre for both the entire grazing season as well as for the summer season after June 30. Native pastures fertilized with ammonium nitrate had the highest carrying capacity.

3. Defoliation disrupts growth regulating systems in seedling plants. At Beltsville, Md., complete defoliation of ladino clover seedlings at several morphological stages reduced subsequent leaf size as much as 60 to 80 percent. If instead of removing the leaves, they were shaded to the photosynthetic compensation point, subsequent leaf size was reduced by only 20 to 35 percent. The remaining two-thirds of the leaf area did not significantly reduce leaf area of subsequent leaves. It appears that defoliation may disrupt growth regulating systems as well as photosynthetic capacity. Leaf growth following defoliation was not influenced by level of stored carbohydrates.

4. Site is of major importance in grass seedling establishment. Near Las Cruces, N. Mex., seedling emergence of black and sideoats grama and lehmann and boer lovegrass was excellent on a sandy loam site rootplowed for creosotebush control. On a silt loam site rootplowed for tarbush control, only a few seedlings of tobosa, alkali sacaton, and sacaton emerged. Seedlings averaged 2 inches tall at the close of the growing season under dead brush on the creosotebush area but were only 1/2-inch tall in open areas. In a basin pitted area from July 29 to September 22, there was an average of 20 days of available soil moisture at a 1/2-inch depth under a brush cover 6 inches from the bottom of the pit. At 36 inches, there was an average of 14 days of available moisture. Without cover, there were only 15 days of available soil moisture 6 inches from the bottom of the pit and 11 days of available moisture at 36 inches. Adjacent flat areas averaged only 8 days of available soil moisture at the 1/2-inch depth. Relatively light dead plant cover reduced soil temperatures up to 41 degrees at a 1/2-inch depth when compared to areas without plant cover.

5. Sand dropseed no longer considered a weed on Southern Plains rangelands. Studies at Woodward, Okla., have shown that sand dropseed and native tall grasses are complimentary to one another. During drought periods yield of tall grasses declined to approximately 80 lbs/acre while sand dropseed increased to 650 lbs/acre. During wet years tall grasses increased to 850 lbs/acre while sand dropseed decreased to 50 pounds. As a result of this complimentary action, range production on sandy soil at Woodward tends to stabilize at around 700 lbs/acre. Sand dropseed is the only important grass on the Southern Plains that increases during drought periods. It should be included in all seedings on sandy soils.

6. Crested wheatgrass equal to native grass in persistence. Crested wheatgrass has proved to be as well adapted to sagebrush sites in southern Idaho as native grasses. Studies of 20- to 32-year old stands show that this



grass has not died out under moderate grazing. It has withstood heat, cold, drought, disease, and heavy grazing well and has been more resistant to fire and severe grazing than native grasses. It has also spread more from volunteer seedlings than native species.

7. Podosporiella verticillata widely distributed in arid soils. The fungus, capable of destroying grass seedlings, has been found to infect seeds planted in southwestern Canada, all of the 11 Western States and North Dakota. The fungus is quite prevalent in the sagebrush-grass vegetation zone and is abundant in the shadscale and mountain brush zones. It has also been found in the Aspen-fir zone in northern Utah. Soils supporting Secale cereale, Bromus tectorum, and other high seed-producing annual grasses have high infection potential.

8. Seed treatment controls Podosporiella verticillata. The fungicides Captan 75, Arasan 75 and Semesan applied to the seed prior to planting resulted in increased germination and seedling emergence in laboratory tests or on native range where Podosporiella verticillata was present in the soil. For spring plantings, 8 ounces of Captan 75 or 6 ounces of Semesan or Arasan 75 per 100 pounds of seed gave good control. Fall plantings, where the seed might be dormant in the soil for several months, required 12 ounces of Captan 75 or 8 ounces of Arasan 75 to control the fungus. Semesan at 6 ounces was not adequate and heavier rates have not been tested. The following genera of grasses are susceptible to the fungus: Agropyron, Bromus, Elymus, Festuca, Sitanion, Secale, and Stipa. Infection has been low on Avena fatua, Phalaris arundinaceae, and Oryzopsis hymenoides. In tests to date infection has not been found on Arrhenatherum elatius. Infection and damage are greatest in early fall seedlings where limited moisture uptake results in slow germination.

9. Brush control improves fair-condition native range. A 40-acre pasture at Burns, Oregon, sprayed with 2,4-D for brush control in 1952 underwent significant species changes but doubled beef production per acre. When yield was adjusted to a median year, squirreltail and Junegrass increased rapidly for 2 years and then gradually declined to below the pretreatment level. Idaho fescue and bluebunch wheatgrass increased slowly for 5 or 6 years, and have not begun a decline; cheatgrass, almost non-existent on the pasture prior to 1956, was, by 1960, the highest producing species, and has since equalled or exceeded bluebunch wheatgrass. Pretreatment total herbage and beef production were 227 and 6.7 lbs/acre, respectively. These amounts have increased, as a result of the 1952 treatment, to 642 and 14.4 pounds, respectively; 89% of the variation in yield was accounted for by fluctuations in the crop-year precipitation.

10. Cactus removal does not affect blue grama production. Cactus was removed from 6 sites on the Central Plains Experimental Range near Fort Collins, Colo., in 1960. Blue grama was clipped from adjacent plots with and without cactus removal on each site from 1960 through 1964. Cactus



removal did not result in increased production of blue grama. On light soils, 22% of the forage in untreated plots was within clumps of cactus and not available for grazing. On heavier soils 15% of the forage was not available.

11. Temperature affects germination of dryland grasses. Under a P.L. 480 project in Israel, it was found that all grass species tested germinated rapidly under 8-hour days at 4°C and warm nights at 20°C. Germination was consistently inferior under reverse conditions. These differences were not accounted for by accumulated degree-hours.

## B. Processing

1. Coastal bermudagrass pellets give highest beef production. In a Coastal bermudagrass utilization study at Tifton, Ga., Coastal bermudagrass pellets gave highest daily gains of 1.88 pounds per day. This compared to 1.81 for hay, 1.36 for continuous grazing, and .84 for green chop. The pounds of beef gains per acre for pelleted Coastal bermudagrass was 788 pounds, which compared to 775 for hay, 600 for continuous grazing, and 529 for green chop.

## C. Management

1. Reed canarygrass gives good beef gains. At St. Paul, Minn., reed canarygrass was compared to brome grass under two grazing pressures and the reed canarygrass pastures did not differ significantly in average daily gains or beef produced per acre from brome grass in 1964. Average daily gains were 1.83 for reed canarygrass and 1.96 for brome grass. Daily gains under heavy grazing pressure were significantly less, 1.78 pounds per day compared to 2.01 pounds per day for light grazing pressure. Beef produced per acre was not statistically different between the grasses or grazing pressures, but trends were toward more beef per acre with reed canarygrass at both grazing pressures.

2. Half-sib animals reduce experimental error in grazing trials. At Columbia, Mo., half-sib and unrelated heifers were again compared during 1964 as a possible method of reducing the animal source of experimental error in grazing trials. It was postulated that the greatest stabilizing effect of related tester animals on the animal source of variation for gain should occur when high quality pasture is available in quantities which will allow each animal to gain at a rate near its genetic potential. Likewise, animal variation in grazing trials should be greatest when average daily gains are lowest. The data obtained support the above assumptions. The half-sib vs unrelated tester comparisons over the past three years suggest that animal sources of experimental error in grazing trials may be reduced by using half-sib testers providing average daily gains on pasture are maintained above approximately 1.2 pounds per day.

3. Nitrate reductase activity related to temperature. At Ithaca, N. Y., studies of nitrate reductase activity in extracts of orchardgrass, brome grass, and timothy showed no levels of nitrate reductase activity at low nitrogen fertilization. Additional nitrogen fertilizer has not led to immediate increases in nitrate reductase activity, rather it appears that

7 to 9 days are required after nitrogen fertilization for the enzyme activity to increase appreciably. Orchardgrass had relatively high nitrate reductase activity when grown at either 60° to 70°F. and with high nitrogen fertility, and a decreased activity at 80°F. Timothy showed highest activity at 70°F., with decreases at either 60° or 80°F.

4. Basic studies indicate that growth rate is regulated by messenger RNA metabolism. In studies at Gainesville, Fla., aimed at the characterization of ribonucleic acid (RNA) metabolism in plants, growth rate is not necessarily correlated with total RNA content or even the level of ribosomes. These results are in contrast to published reports which show a positive correlation between growth rate and total RNA and ribosomal contents. In these studies, growth rate appears to be regulated either by messenger RNA formation or attachment. Pulse-type studies with P<sup>32</sup> indicate that the polysomes which form in the faster-growing plants do in fact contain greater amounts of the isotope than the slower growing tissues.

5. Photosynthetic difference as shown in cool season vs warm season grasses. At Beltsville, Md., the effect of temperature on photosynthesis of pangolagrass (warm season) was quite different from the effect of temperature on photosynthesis of orchardgrass (cool season). At 10°C. the photosynthetic rate of orchardgrass was twice that for pangola. Orchardgrass reached its maximum photosynthetic rate of 3.6 mg. CO<sub>2</sub>/cm<sup>2</sup>/min. at 20 to 25°C., and the photosynthetic rate decreased to 2.2 mg. CO<sub>2</sub>/cm<sup>2</sup>/min. at 30 to 35°C. Pangolagrass, on the other hand, showed a continuous increase in photosynthesis with increasing temperature. Photosynthetic rates for pangola at 20 to 25°C. and 30 to 35°C. were 6.0 and 7.7 mg. CO<sub>2</sub>/cm<sup>2</sup>/min., respectively. Low temperatures were more critical for pangola and high temperatures were more critical for orchardgrass, in terms of photosynthetic response.

6. Weather data used to predict growth of cereals. At Tifton, Ga., regression equations were developed for forage yields of winter cereals using environmental factors, including temperature, solar radiation, and precipitation. The best-fitting equations accounted for 96, 91, and 93 percent of the variations in yield of Arlington oats, Wrens Abruzzi rye, and Bledsoe wheat. These equations and weather data for the past 40 years were used to calculate the theoretical frequency distributions of forage yield for each month during winter and spring.

7. Nutrient cultures show deficiency symptoms and response to sulfur fertilization. Carefully controlled nutrient culture studies at Berkeley, Calif., demonstrated symptoms of sulfur deficiency in Italian ryegrass and Spanish clover. Yield responses to S fertilization were also shown. S deficiency in Italian ryegrass produced general chlorosis with young blades first showing a pale lemon-yellow color, yellowing of the entire upper-blade surface with a pinkish-red color along the blade edges and under-surface of younger blades, a more than normal erect habit of growth, a



reduction in length of internodes, and fewer fibrous roots but roots larger in diameter than those of nondeficient plants. Symptoms of S deficiency on Spanish clover were similar; but with severe S deficiency, brown spots were scattered at random over the upper and lower surfaces of the young leaflets. With an increase in S fertilization from .01562 to 1 mg. S per liter of solution on both Italian ryegrass and Spanish clover, dry weight of tops increased 4.9-fold and 4.5-fold, respectively; dry weight of roots increased 1.6-fold and 1.1-fold, respectively; and total dry weight of tops plus roots per unit of sulfur added decreased from 1,900 to 100 mg. and from 1,990 to 110 mg., respectively.

8. Stocking rate and beef production influenced by soil. Studies at Woodward, Okla., over the past 20 years reveal gains per head under proper stocking to be relatively constant but gain per acre is strongly influenced by soil texture, particularly silt and clay content. These studies have shown proper stocking per section, and gains per acre to be as follows: Summerlong on Tivoli dune sand, 94 steers and 44 lbs. of beef/acre; Pratt loamy sand 116 steers and 57 pounds of beef; Pratt sandy loam 160 steers and 76 pounds of beef.

9. Alfalfa can be maintained in crested wheatgrass-alfalfa pastures. At Mandan, N. Dak., in a management system where crested wheatgrass-alfalfa pastures were grazed only in the spring for a period of 35 to 40 days and not beyond July 1, alfalfa maintained itself or increased in the mixture. When such a mixture was grazed season long, spring and fall grazed, or spring and summer grazed, alfalfa disappeared. Mixtures established in 1954 and spring grazed each year for 9 years contained 10% alfalfa by weight in 1956 and 52% in 1964. Crested wheatgrass-alfalfa mixtures were superior to straight crested wheat-grass in all measurements of pasture production. For the 9 years, a comparison of straight crested wheatgrass with the mixture showed gains per acre to be 101 and 131 pounds, daily gains 2.64 and 2.79 pounds, steerdays grazing 38 and 52 per acre, and TDN 380 and 482 pounds per acre, respectively.

10. Monthly forage production of native range studied on Southern Plains. Sixteen years' data at Woodward, Okla., reveal that there is tremendous seasonal variation in monthly forage production. The range in dry matter production has been from 70 to 500 lbs/acre in May; 30 to 420 in June; 20 to 580 in July; 0 to 680 in August and 20 to 540 in September. On the average, during July and August, forage production is 30% higher than during May and June.

11. Proper grazing intensity important on Northern Plains rangelands. A 12-year study at Miles City, Mont., showed the cumulative effects of too many cattle per unit of land caused serious reduction in fertility of beef cows and growth rate of calves. Range forage production was reduced under heavy grazing. The more palatable and productive forage species decreased and the undesirable species increased. Carrying capacity was greatly



reduced during drought. Undesirable soil characteristics influencing soil moisture storage occurred under heavy grazing, and there was an increase in the cost of supplemental winter feeding and an increase in death losses.

12. Protein supplement effective in winter grazing management. Quality of pasture determines level of protein feeding for best winter gains according to studies conducted at Woodward, Okla. Three-year averages for winter gains per steer on sorghum pasture were, with no supplemental protein, 96 pounds per head; with 1-1/2 pounds cottonseed crumbles (41% protein) daily, 194 pounds per head; and with 3-1/2 pounds crumbles daily, 215 pounds per head. On sorghum pasture 1-1/2 pounds crumbles daily was highly profitable. In contrast, steers on low quality Caucasian bluestem winter pasture gained 50 pounds per head when given 1-1/2 pounds crumbles daily and 120 pounds per head when given 3-1/2 pounds daily. In a study during the winter of 1963-4 a constant rate of feeding protein supplement appeared better than a progressively increasing rate, although gains did not differ significantly.

13. Amount of forage removal determines profit. A 23-year grazing study near Nunn, Colorado, has indicated the proper amount of annual forage removal during the warm season. The removal of approximately 40% of the perennial range forage each year has resulted in the maintenance of normal forage production, reasonable cattle gains, and higher monetary returns per acre than either 20% or 60% removal. Heavy use of rangeland for 23 years did not alter species composition appreciably, but it did lower production. After a few years of rest from heavy grazing, blue grama produced as much as that on lightly used range.

14. Effects of "boss order" studied among weaner steers. Seven-day effects are noticeable, but monthly weighings show only insignificant differences in gains of steers. A 3-year study at Woodward, Okla., gave average year-long gain per head of 460 pounds from continuous grazing, 456 pounds when the steers were moved to a new pasture (same total area grazed) each month, and 452 pounds when steers were both rotated monthly and put with strange associates. Weighed 7 days after changes in environment the 3-year average gains were 9, 4 and 6 pounds, respectively. Rotation and "boss order change" appear to have an early (7-day) effect which is essentially overcome by the end of the month.

15. Technique for more accurate cattle weights. At Woodward, Okla. where it takes 3 to 4 hours to obtain initial cattle weights for grazing experiments, a technique was developed to obtain more accurate weights. Animals weighed first have no shrink, whereas those weighed later may have shrunk up to 4 hours. The problem was corrected by holding the first 20 steers in a separate pen and re-weighing them at 2-hour intervals to obtain an accurate constant to be used to correct shrunk weights.

#### D. Quality and Varietal Evaluation

1. Decline of forage quality, a result of changes in stem rather than leaf

composition. At Lafayette, Ind., in vitro digestibility estimates were obtained on plant parts of alfalfa and reed canarygrass. The whole plant tissue of alfalfa declined 0.40 digestibility units per day while reed canarygrass declined 0.50 units per day. In both species stem tissue initially had the greatest digestibility, but declined at a much more rapid rate than leaf tissue with advancing maturity. Stem tissue and sheath tissue of grass from the upper portion of the plant was greater in digestibility than the lower portion. The sheath tissue was lower in digestibility than stem tissue from the same stem segment. Leaf tissue of alfalfa essentially remained constant over the 30-day growth period from approximately 1/10 bud to seed pod formation. The leaf tissue of reed canarygrass declined with maturity but at a slower rate than the corresponding stem and sheath tissue. The digestibility of leaf tissue exceeded stem and sheath tissue only after head emergence.

2. Cellulose fraction of the cell wall has higher digestion coefficient. At University Park, Pa., chemical studies on the major constituents of the cell walls of forage plants, hemicellulose, cellulose, and lignin showed that the grasses contain more hemicellulose and less lignin than alfalfa. The hemicelluloses are less digestible by ruminants than celluloses in all of the forage studies. Lignin is more closely associated with hemicellulose than it is with cellulose, and this is particularly true in alfalfa. This leads to the conclusion that the cell wall is not digested as a unit by ruminants but that the hemicellulose and cellulose are digested independently.

3. Fecal indicators useful in predicting pasture digestibility. At St. Paul, Minn., certain fecal indicator predictors such as chromogen and nitrogen were sufficiently accurate to be used to predict digestibility in pasture studies. Using sheep, these two indicators were correlated and equally sensitive statistically as predictors of digestible organic matter in pasture forages. When used in conjunction with fecal output to measure intake, the chromogen and nitrogen methods were equally accurate. When fecal output was combined with digestibility estimates and measure of intake and digestibility, organic matter was obtained. Both methods yielded an estimate of 2.1 pounds of digestible organic matter consumed per sheep per day average for all forages tested. This compared favorably to the 2.2 pounds TDN computed to be required per sheep based on feeding standards.

4. Russian wildrye superior to native range for late fall grazing. Three-year results at Mandan, N. Dak., have shown that Russian wildrye pasture can extend the fall grazing season, an important need of Northern Plains livestock operators. Yearling steers were fall grazed on pastures of Russian wildrye and native grass that had been held all summer without grazing. The average grazing intensity was .62 and 1.38 head/acre, respectively. Steerdays grazing/acre were 84 on the Russian wildrye and 37 on native. Gains/head were 25 and .7 and gains/acre were 40 and 0, respectively. Production and consumption of dry matter were also much higher on Russian wildrye.



5. Sheep used to determine digestibility of meadow hay. Wether sheep in digestion crates at Burns, Oreg., were fed meadow hay harvested at weekly intervals from June 21 to August 9. Digestibility of measured constituents remained fairly constant through late June and early July and then decreased throughout July and early August. Digestibilities of nitrogen, dry matter, cellulose, and gross energy were 64, 65, 72, and 64%, respectively, for hay cut June 21; and 40, 51, 60, and 50%, respectively, for hay cut August 9. In another trial using meadow hay fertilized with various rates of nitrogen applied on four different dates, there was no measured affect of the fertilizer on digestibility of nitrogen, cellulose, or dry matter.

6. Quality of range forage influenced by precipitation. In long-time studies at Woodward, Okla., winter gains of steers were higher on lightly grazed pastures (adequate feed) under low than under high summer or fall precipitation. Following dry summers (11 inches average precipitation), winter steer gains averaged 105 lbs/head while following wet summers (26 inches average precipitation) only 85 lbs/head. When fall precipitation averaged only 1 inch, winter steer gains were 90 lbs/head. In years averaging 8 inches fall precipitation, gains were 65 lbs/head. Summer gains were not so markedly influenced by concurrent precipitation, but calf weaning weights averaged 7 lbs/calf higher in wet summers.

7. Nitrate content of irrigated Russian wildrye affected by row spacing and fertility level. Forage from irrigated rows of Russian wildrye at Mandan, N. Dak., was harvested at weekly intervals from May 2 to July 13. The nitrate level of the forage increased as the row spacing increased from 6 to 18 to 36 inches. It also increased at all row spacings as the rate of nitrogen fertilizer was applied at rates of 0, 100, 200 and 400 pounds/acre. On May 2, the nitrate contents of the forage from the 6-, 18-, and 36-inch rows with 0-N were .053, .066 and .176, respectively. At the 400 pound N rate the nitrate contents were .304, .902, and 1.241%, respectively, all above the .3% level sometimes considered the maximum to be safe for livestock. The nitrate content decreased with advancing maturity. On June 13, after flowering, only forage from the 400 pound N level for the 36-inch spacing was above .3% nitrate.

8. Weeping lovegrass hay a good winter feed. Weaner steers made better winter gains from weeping lovegrass hay at Woodward, Okla., than from winter grazing of summer-deferred weeping lovegrass range, or from winter on year-long grazed native range. Steers on weeping lovegrass hay and aftermath during winter gained 77 pounds/head and produced 89 pounds of beef/acre. On summer-deferred weeping lovegrass range, winter gains were 12 pounds/head and 9 pounds/acre, while on native range, gains were 69 lbs/head. When native range is summer deferred, steers usually produce 25 to 30 pounds beef/acre during the winter grazing season.



9. Techniques for polyfructosan analysis developed. Progress on this PL 480 project in India to date has been largely the development of a technique for polyfructosan analysis. That changes in polyfructosan composition occur with stages of maturity may help explain the changes in forage quality associated with different stages of maturity.

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SEED CROP CULTURE, DISEASES, PHYSIOLOGY, NUTRITION,  
HARVESTING, AND VARIETY EVALUATION  
Crops Research Division, ARS

Problem: Domestic production of many grass and legume seed crops is well below national consumption. Importation of seed is necessary because technology has not been developed to make seed production a profitable farm enterprise. Research is needed to investigate the factors that limit tiller initiation, fertilization and seed formation, and to develop cultural and management techniques including disease control practices that will assure efficient seed production. Fundamental physiological investigations on growth responses of grasses and legumes to various environmental, management and micro-climate factors are needed to establish the specific requirements for reproduction. Normal vegetable seed supply is adequate, but geographic concentration of the seed industry is potentially hazardous. Mechanization of the vegetable industry requires increased emergence and uniformity of seedling development, but improvements in processing quality of vegetables often decrease seed quality. There is insufficient basic knowledge of physiology of seed development and germination as these influence the crop-producing potential of seeds and potential industry mechanization.

USDA AND COOPERATIVE PROGRAM

Grasses and Legumes. The Department has a continuing long-time program involving agronomists, physiologists, and pathologists engaged in both basic and applied research. Investigations are underway at Shafter, Calif.; Lafayette, Ind.; Stillwater, Okla.; Corvallis, Ore.; Logan, Utah; and Prosser and Pullman, Wash. All work is conducted in cooperation with the respective State Agricultural Experiment Stations and the Entomology and Agricultural Engineering Research Divisions.

Research is being conducted under three P. L. 480 contracts-- one in Finland, one in Israel, and one in Germany. Studies under the three projects are co-ordinated with the broad research objective of investigating the influence of environment and management practices on population shifts in cross-pollinated forage-crop varieties when seed is produced in areas outside the regions of origin.

The Federal research in this area totals 13.5 professional man-years. Of this number 4.8 are devoted to culture, 4.2 to physiology, 1.0 to diseases, and 3.5 to variety evaluation.

Vegetable Crops. Basic research on physiology and biochemistry of vegetable seed germination and seedling vigor is carried out at Beltsville, Maryland. At Logan, Utah, in cooperation with the State Agricultural Experiment Station, studies involve seed quality as controlled by environmental conditions, including cultural and harvesting practices. Beltsville

and Logan programs are closely co-ordinated.

In India, under P. L. 480 projects, research includes physiology of development of ovules and other reproductive organs and early production of ascorbic acid and ascorbic acid oxidase and the metabolism of bound forms of nicotinic acid (niacinogen) in germination. In Israel, sub-microscopic structural changes in mitochondria and other cellular structures are studied in close cooperation with Beltsville scientists.

#### PROGRAM OF THE STATE EXPERIMENT STATIONS

Scientists at the State Experiment Stations are engaged in basic and applied research in physiology, pathology, agronomy and engineering pertaining to seeds. In several States, the research is conducted cooperatively with the Department. This research is continuing to provide useful fundamental information for the improvement of seed crop culture, harvesting, and storage. Two regional projects have been organized by the States to coordinate research on seed problems. In the northeastern region under NEM-22, chemical, physical, physiological, morphological, pathological, and other means of determining varietal purity of seeds are being sought. In the Western region under WM-35, much attention is being given to developing and improving techniques for rapid estimation of viability of seed, factors during growth affecting viability, harvest and storage factors relating to poor germination, and improved laboratory methods of determining the viability of and purity of range grass seed. Other studies concern management of seed production fields for forage seed, control of insects, effect of growing conditions on seed quality, harvesting equipment, and drying practices. Seed storage research concerns factors associated with seed deterioration, the sequence of physiological changes, storage factors affecting these changes, seedborne microflora, and the use of seed coatings. Testing procedures for germination, vigor, and stand-producing potential are receiving much attention. Some work is being done on blending seed for uniformity. Various means of breaking seed dormancy including radio-frequency electric fields are being explored. In addition to studies on seed production and seed technology, most of the States are involved to some degree in producing breeder, foundation and certified seed of newly developed crop varieties.

Disease problems of the major seed crops of vegetables, ornamentals, forages, and field crops are being studied. Seed-borne diseases are known to limit production. In addition, there is an increased demand upon seeds to provide a plant that meets exact requirements for culture, harvesting, processing, and product quality. The need for knowledge on disease problems has increased. Some of the research in progress is designed to provide new knowledge on specific causal agents of disease. In other projects,

scientists are concerned with isolating the components of resistance to disease, so that these may be used by plant improvement specialists and others to provide resistant plants of commercial value. A number of projects are designed to provide the critical histological and histochemical evidence that is essential to the study of certain seed crop diseases. The role of fungi and bacteria in the deterioration of seeds is being emphasized in some projects.

The total research effort in this area of research is 19.1 professional man-years - of which 1.4 is for culture, 0.5 for diseases, 3.3 for physiology, 1.0 for harvesting, and 12.9 for variety evaluation.

## PROGRESS--USDA AND COOPERATIVE PROGRAMS

### A. Culture

1. Reproduction in red clover clones is influenced by environment. In cooperative studies at Lexington, Ky. (38° latitude) and Prosser, Washington (46° latitude), the reproduction characteristics of 30 red clover clones were investigated. Characteristics most nearly constant at both locations were number of seeds per head and number of seeds per floret. Slightly more florets per head were found at Prosser than at Lexington, but the greatest difference was in the large number of heads produced at Prosser which resulted in higher seed yields. The significant interaction of clones by locations, particularly in date of first bloom and number of heads in bloom at different times throughout the season, provided an opportunity for nonrandom pollination. This may contribute to shifts in gene frequency and subsequent loss of adaptation in the forage-producing area. Seed yield at both locations among polycross progenies was less than among the parental clones. Lack of correlation between seed yield of clones and their progenies indicates that seed yield of vegetative transplants should not be a criterion for selection of genotypes for a synthetic variety. Progeny testing may be necessary to insure that a synthetic variety possesses high seed-yielding potential both inside and outside its area of adaptation.

2. Effect of processing native grass seed on quality and stand establishment. Processing chaffy range grass seed, varying from slightly trimmed to clean caryopses, was studied at Stillwater, Okla. Results indicate that processing to the extent of extracting the caryopses damages seed quality. Freshly extracted caryopses usually germinate faster and more completely in laboratory tests than caryopses which are left in their enclosing appendages. However, slight mechanical injuries reduce viability and stand establishment under field conditions. In general, with the exception of buffalograss seed burs, unprocessed seed and that which had only a portion of its appendages removed performed best under field conditions.

3. Cultural methods, fertilizers, and herbicides influence seed yields and quality of Kentucky bluegrass. In the fourth crop year at Corvallis,



Oregon, seed yields of Newport Kentucky bluegrass planted in 30-inch rows were 75% of those in solid stands. Lime had no significant affect on seed yield, but there was a slight lime-nitrogen interaction. Yield differences due to nitrogen, sulphur, and cultural methods were highly significant as were nitrogen cultural method interactions. Heavy spring nitrogen applications stimulated spring growth and seed yields. Treatment with diuron controlled weeds and increased seed yields significantly.

4. Seed quality is critical factor in establishing stands. At Lincoln, Neb., differences in seed quality as well as yield were found in side-oats grama seed produced under different management practices. Seed size was highly correlated with seedling vigor. The proper number and time of irrigations were important. Seed produced with two irrigations--one in fall and a second at time of heading--had a seed quality of 84% while that produced with only one irrigation in the fall had a quality of 48%. Nitrogen fertilization along with adequate soil moisture influenced seed set and quality. Three-year average yields of pure seed per acre as well as seed set and seed quality were best from 40 pounds of nitrogen per acre. Applications of 80 pounds of N. per acre gave some increased seed set and yield as stands became older. Use of good seed production practices increased number of seeds per unit weight of spikes from 132 to 358 and quality from 23% to 72%.

5. Cross-pollination among alfalfa varieties may be associated with species of pollinators. At Logan, Utah, honeybees were more efficient cross-pollinators than leaf-cutting bees, but seed production was higher for the latter. Seed production of a white-flowered alfalfa was independent of the purple-flowered variety with which it was paired. Varieties produced seed in relation to their genetic potential. Seed yield was not affected by pollinator-variety interactions, but cross-pollination was affected. The white-flowered alfalfa was equal to colored flowered alfalfa in attractiveness to nectar-collecting honeybees, but it was less attractive to leaf-cutting bees. The white- and colored-flowered alfalfas did not differ significantly in percentage of stainable pollen, nectar sugar per flower, or tripping velocity as measured by pollen extrusion.

6. Germination of normal and hulled grass seed is affected by storage conditions. At Pullman, Wash., normal seed of six cool-season grasses stored cold under low humidity showed no significant reduction in germination during a five-year period. Orchardgrass and meadow foxtail seed was not reduced in germination by warm storage but was lowered by cool storage associated with high humidity. Germination of both intermediate wheatgrass and smooth brome grass was reduced in both warm and cool storage. Timothy and tall oatgrass seed retained germination for the five-year period regardless of storage condition. Hulled seed of tall oatgrass, orchardgrass and brome grass retained essentially the same germination as the normal seed under cold storage conditions; whereas, hulled seed of timothy, intermediate wheatgrass, and meadow fescue suffered a significant decrease. Hulled seed of all six species was significantly reduced in

germination by warm or cool storage as compared to cold storage with low humidity.

7. Seed set in red clover may be directly related to nectar volume.

Measurements of nectar and floral characteristics of red clover were studied at Prosser, Wash. Seed yield data suggest that attractiveness to pollinators is related to nectar volume. During a five-day interval, nectar volume increased sharply to the sixth day. Sugar concentration did not vary significantly. The average number of seeds per floret when heads were exposed from the third to the seventh day were: .27, .33, .34, .42, and .19, respectively. Decrease in seed set on the seventh day coincides with the observed decrease in nectar volume in florets.

8. Distribution of foundation seed of forage species. Distribution of seed of superior forage-crop varieties by the Foundation Seed Project amounted to 238,391 pounds in 1964. This was a decrease from the previous year because of limited supplies of Vernal alfalfa and Kenland and Pennscott red clovers. Foundation seed was distributed to 13 States and Canada. A total of 459,289 pounds of foundation seed was produced under CCC contracts in Arizona, California, Idaho, Nevada, and Washington. The supply of each variety is adequate to meet expected demands in 1965.

B. Physiology

1. Temperature relations in bean germination. In Beltsville, Md. research, the earliest stage in imbibition of lima bean seeds and excised embryonic axes is temperature sensitive. Imbibition at 60°F or lower reduces subsequent growth at 76°; imbibition at 76° protects against subsequent chilling injury. Low vigor (bleached) seeds are more sensitive to injury than high vigor green seeds. Seed responses can be variable because the seed coat may restrict water uptake permitting the seed to avoid chilling injury. This protective mechanism is most effective at low temperature under relatively dry conditions. Thus the total temperature sensitivity is complex and can result in extremely variable seed germination. Snap bean varieties released prior to 1955 are not susceptible to chilling injury, but many newer varieties can be extremely sensitive, even to 68°F, a normal germination temperature.

2. Germination of grass seed improved by special treatments. Seed dormancy inhibits germination in seed of some grasses. Studies with Beaked panicum, Panicum anceps, at Stillwater, Okla., showed that seed dormancy can be broken by soaking seeds for 6 to 8 hours in concentrated sodium hypochlorite or by prechilling 6 to 8 weeks at 5-10° C on moist substrate. The beneficial action of sodium hypochlorite on germination is evidently through destruction of the outer glumes and possibly scarification of the inner seed coat. As seeds age, the degree of seed dormancy becomes variable and less pronounced. The prechill requirement, whether continuous or broken under optimum conditions, initiates germination. Respiration studies revealed that oxygen uptake by seed increased rapidly between the

fourth and fifth week of prechill. The amount of carbon dioxide evolved increases with each additional week of prechilling. At six weeks the total amount of carbon dioxide released was twice as great as that recorded for the first week.

3. Photoperiod affects dryweight accumulation in forage species. At Lafayette, Ind., dry matter accumulations of four legumes and three cool-season grasses were determined at 15, 20, 25, 30, and 35° C, 12- and 18-hour photoperiods, and high light intensity. Dry matter accumulation under a 12-hour photoperiod for all species was about 50% less than comparative accumulations at 18-hour photoperiods. No differences among the various species were observed. The 12-hour photoperiod, however, provides only two-thirds as much photosynthetic energy as the 18-hour photoperiod. This suggests that dry matter accumulations which are in excess of the expected growth reduction due to reduced energy levels represent fundamental changes in plant metabolism. Responses of individual species to the five temperature regimes, except for reduced dry matter accumulation, were the same at the 12-hour photoperiod as for the 18-hour photoperiod.

4. Flower production, seed set and seed yield of alfalfa are influenced by temperature. In a study at Prosser, Wash., clones of Ferax alfalfa were grown in constant and alternating temperature regimes. Under constant temperatures of 21 to 29° C seed yields decreased with increasing temperature. When night temperatures were held at 21°, seed yield also decreased with increasing day temperatures. When night temperatures were 14° and day temperatures from 14 to 20°, seed yields were not significantly different. Lower seed yields at higher temperatures were correlated with production of fewer pods and fewer flowers per raceme. Flowering and pod maturation were observed to occur in cycles. As the number of pods on a stem increased, rate of terminal growth decreased. Eventually the terminal bud died. This led to the hypothesis for alfalfa that the primary condition limiting pod production, and therefore seed production, is capacity of the plant to supply metabolites to buds and to ripening pods in the required quantities.

5. Specific techniques will differentiate alfalfa ecotypes. At Lafayette, Ind., polyacrylamide gel electrophoresis of cytoplasmic root proteins was used to distinguish between alfalfa genotypes. From 18 to 21 protein bands were found depending upon genotype and physiological condition (cold-hardened or non-hardened). A general electrophoretic pattern was observed for all samples, but there were enough variations within the patterns developed by the six genotypes to allow identification in each physiological condition. One specific band was generally more prevalent in the hardened than in the non-hardened condition. Serological analyses confirmed that there were antigenic differences between genotypes and physiological conditions.



6. Submicroscopic cellular structural changes in germination. Under P. L. 480, preliminary studies in Israel on the relationship between temperature sensitivity of lima bean seeds and submicroscopic structure suggest that in the dry embryonic axis the endoplasmic reticulum is oriented near the cell boundaries, becoming randomly oriented during early imbibition and early growth.

7. In vitro fertilization of excised ovules. In P. L. 480 research at Delhi, India, although pollen germination proceeded readily, ovules of Zephyranthes Lancasteri excised and inoculated in vitro were not penetrated by pollen tubes, thus suggesting the necessity of stimuli to orient pollen tube growth toward the ovule.

#### C. Diseases

1. Aerated steam treatment for seed disinfection shows promise. At Corvallis, Oregon, treating seeds with aerated steam gave excellent eradication of a variety of fungi carried on the surface of and internally in red clover seed. This method shows promise for disinfection of limited quantities of seed that will serve as a nucleus for commercial increases. Tilletia pallida smut was eliminated from Kingston velvet bentgrass by the aerated steam treatment and made possible release of this variety.

#### D. Variety Evaluation

1. Growth responses from seed of several forage-crop species produced in different regions. At Lafayette, Ind., studies were conducted to evaluate the vegetative and floral characteristics of brome grass, orchardgrass, timothy, birdsfoot trefoil, red clover, white clover, and alfalfa progenies produced in different environments at Shafter and Tehachapi, Calif.; College Station, Texas; Logan, Utah; and Prosser, Wash. To statistically analyze these data, a program capable of performing the analysis of variance for balanced or unbalanced lattice, lattice square, rectangular lattice, triple lattice, and repetitions of these designs has been modified and made operational for a 7094 computer. Previous experimental analyses indicate upwards of 10 to 25 percent increase in efficiency with lattice designs at Lafayette.

2. Influence of environment on population shifts in clover varieties. Investigations under a P. L. 480 project in Finland indicate only slight population shifts occurred when seed of Finnish alsike, red and white clover varieties were produced in the United States and Canada. Evaluation tests indicate commercial seed increases of these varieties can be made in the U. S. for one or more generations without significant population shifts.

3. Influence of environment on genetic shifts in forage-crop varieties. P. L. 480 projects have been initiated in Israel and Germany to study the effects of temperature, photoperiod, and management on plant ecotypes constituting several grass and legume varieties. Seed of forage grasses and legumes developed in the United States, France, and Finland were multiplied

in Israel. These first-generation progenies were established in evaluation tests.

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WEED AND NEMATODE CONTROL  
Crops Research Division, ARS

Problem. Weeds cause losses in crops, orchards, grazing lands, forests, water supplies, and irrigation and drainage systems. These losses can be reduced by finding more effective chemical, biological, mechanical and combination methods of weed control.

Plant-parasitic nematodes occur in all soils used for growing of crops and attack all kinds of plants grown for food, forage, fiber, feed or ornamental purposes. Severity of attack by certain fungi is increased if nematodes are present. Nematodes also have been known to be the vectors of several plant viruses. There is need for improvement in methods of controlling nematodes on grain and forage crops.

USDA AND COOPERATIVE PROGRAM

Much of the weed and nematode control research in the Department is cooperative with State Experiment Stations, other Federal agencies, industry and certain private groups; and is cross commodity in nature. The total federal weed control program involves 78.2 professional man-years' effort. Of this total, 4.0 man years are specifically directed to weed control in grain crops, 1.2 in rice; and 23.0 in forage and range plants. The total federal nematode control program involves 27.3 professional man-years' effort of which 0.1 are devoted to cereals and 1.2 to forage and range crops. There are P.L. 480 projects at Poznan, Poland, to study the effects of chemical and mechanical control methods on weeds, corn, and associated crops and at Lucknow, India, for biochemical studies on angiosperm parasites--dodder and witchweed.

PROGRAM OF STATE EXPERIMENT STATIONS

All the State experiment stations are conducting basic and applied research in weed control. These studies involve evaluation of selective herbicidal properties of new chemicals to show the relation between chemical structure, herbicidal activity and weed-crop selectivity; the nature, behavior, and effect of herbicides on their degradation products in and on plant and plant products; the mechanism of herbicidal action; influence of climate, plant morphology and soil characteristics on the effectiveness of herbicides in selectively controlling weeds and on their persistence in plant tissue. Studies are being conducted on the movement and persistence of herbicides in various soil types and the phenomena involved in absorption and other interaction of herbicides with clay complexes.

Weed life cycles and growth habits are being studied under different environments to determine the most susceptible stage of vulnerability to

herbicides and other control measures. Other aspects that are currently being investigated are: competition between weeds and desired plant successions following control measures including replacement vegetation and management practices. Relation between weeds and biological control organism that attack them in different environment is being studied on a limited scale.

Much of the basic research in weed control is being done via six regional projects as follows: W-52 is exploring the fundamental biochemical and biophysical processes involved in herbicidal action; W-63 is studying the chemical and physical properties of herbicides in relation to environment and effectiveness; NE-42 is investigating weed life cycles and light as factors in weed control; NC-61 is concerned with the nature and extent of competition between weeds and crops; S-18 and NE-42 are investigating the behavior of herbicides in soil, the physiological aspects of certain herbicides and life histories of important southern and northeastern weed species. CFR-1 program is attacking basic problems in aquatic weed control and brush control. The USDA cooperates on much of this research activity.

The total State scientific effort devoted to weed control research is 357.0 professional man-years.

Nematode investigations are being conducted at most of the State Stations and many of these scientists participate in the four Regional Research Projects concerned with phytonematology. Through these and other projects at the various institutions, scientists are contributing new knowledge on the genetics, physiology, and pathology of nemas. Some station scientists, as a result of their recent findings on nemas as vectors of viruses, are conducting intensive investigations of the biologies of this process. Other research on fundamental problems in nematology as well as work on identification and control are indicated in the appropriate crop section of this report.

The total research effort on nematode identification, physiology, and control at the State Stations is approximately 54.9 professional man-years.

#### PROGRESS - USDA AND COOPERATIVE PROGRAMS

##### A. Weed Control

1. Grain Sorghum. Regardless of application technique, atrazine caused greater initial retardation of grain sorghum than did 2-chloro-4,6-bis (isopropylamino )-s-triazine (propazine) in Kansas. Preemergence propazine or preemergence atrazine was better for weed control in grain sorghum than were other pre- or postemergence treatments. Atrazine applied after harvest of wheat in 1963 combined with two cultivations of grain sorghum in 1964 was better than either chemical treatment alone or cultivation alone as a

weed control practice in a wheat-sorghum-summer fallow rotation.

Picloram, reported promising in 1963, gave outstanding control of field bindweed when applied at 3 to 4 lb/A in Kansas. No new herbicides appeared outstanding.

2. Witchweed Control in Corn. An unidentified alkali-labile corn root exudate stimulates germination of witchweed. Raising the  $p^H$  of an acid soil to 8.0 resulted in decreased witchweed emergence and increased vigor of corn in North Carolina. A 32% separation of witchweed seed from soil was obtained with a recently developed electrostatic seed separator. Up to 89% recovery from soil was obtained with a device used in State seed testing laboratories. Research on the direct control of witchweed in corn with herbicides and indirect control in nonhost crops through herbicides control of host weeds continued to result in normal or increased yield of crops. Season-long control of witchweed and of witchweed host plants was obtained with several herbicides and several combination treatments.

3. Annual Weed Control in Corn. In Mississippi, a new substituted urea, 2,4-D, and 2,4,5-T incorporated in the soil gave good weed control in corn without crop injury. Atrazine incorporated at 4 lb/A resulted in serious crop injury.

In a P.L. 480 project in Poland, it has been found that there was a strong influence of triazine herbicides on the sugar content of corn, largely a drop in saccharose and fructose. This may lead to a new concept of the mechanism of action of triazine herbicides.

4. Rice. In Arkansas, rice was significantly injured if propanil was applied too soon (15 days) before or after application of parathion. Application of nitrogen to rice 10-15 days before treatment with 2,4-D or 2,4,5-T increased the rice injury from the herbicides. Invert amines of phenoxy herbicides injured rice more than regular amines.

Methyl thiomethyl 2,3,5,6-tetrachloroterephthalate, ethyl-1-hexamethylene=iminecarbothiolate, 3,4-dichlorobenzyl-N-methylcarbamate, potassium azide, picloram, and IPC were promising new materials.

Curly indigo competition for 116 days following emergence reduced rice yields by 9 to 26 bu/A, and competition from barnyardgrass in excess of 40 days after emergence reduced yields by 18 to 89 bu/A.

5. Weed Control in Grass Crops Grown for Seed Production. From 15 to 20% of speciality grasses planted for seed fail to become established because of weed competition. Less than 10% of the established fields produce seed that can be certified the first year due to presence of weeds. In Oregon, chemical seedbed treatments followed by postemergence treatments, promise



to largely eliminate both of these problems. 1,1'-dimethyl-4,4'-bipyridinium salt (paraquat), simazine, atrazine, prometryne, picloram, and diuron are the herbicides most promising in the seasonal program for grasses.

6. Dodder Control in Alfalfa. Practical full season dodder control in Washington in first and second crop alfalfa resulted from soil applications of CIPC, butynyl N-(3-chlorophenyl)carbamate (BIPC), dichlobenil or DCPA in combination with tillage. Several new herbicides and combinations of herbicides had a high degree of dodder-killing activity in initial trials.

## 7. Weeds in Grazing Lands

a. Ecological Studies. In experiments at Lafayette, Indiana, the most important competitive factor in the establishment of forage legumes was found to be light when moisture supply is adequate. Light competition retards the vegetative regrowth potential by reducing the development of crown buds and shoots of birdsfoot trefoil. In the early stages of establishment, broadleaf weeds are more competitive than weed grasses. Light competition from increased weed density is not as important as competition for moisture in the soil. Also, roots from weed grasses such as yellow foxtail, giant foxtail, and crabgrass appear to contain substances which are inhibitory to the growth of corn roots. Of the three grasses mentioned, giant foxtail contains the greatest amount of inhibitor, if all three contain the same substance, or the most toxic if they are different.

b. Physiological Studies. A method for determination of phenoxy herbicides in forage plants has been improved in research at Ithaca, New York. Basically, the technique involves determination of the herbicide by electron affinity gas chromatography after extraction and improved liquid-liquid phase separations for cleanup. The method is good for 4-(2,4-dichlorophenoxy)butyric acid (2,4-DB), 2,4-D and silvex in alfalfa, red clover, brome grass, timothy and orchardgrass. Birdsfoot trefoil presents minor difficulties because of a tendency to form emulsions.

Limited simulated rainfall appeared to enhance absorption of 2,4-DB in birdsfoot trefoil, alfalfa, brome grass, orchardgrass, and timothy. A definite period of time seemed to be necessary for the plant to metabolize absorbed herbicide regardless of a rainfall variable. Partial degradation of 2,4-DB, 2,4-D and silvex apparently takes place in ensilage of orchardgrass, brome grass, and timothy. On occasion, phenoxy herbicides in legume silages were not degraded. The reason for this is presently unknown. In the majority of cases, there was significant degradation of phenoxy compounds in legume silage.

The alkaloid content in the leaves of tall larkspur (Delphinium occidentale) and false hellebore (Veratrum californicum) was significantly increased following treatment with 2,4,5-T. Three species are known to be thus affected by this herbicide. Also at Logan, Utah, biochemical work on isolating and identifying the toxic substance from timberline milkvetch (Astragalus miser) proceeds slowly, but some progress has been made.

8. Weed Control in Pastures. A new herbicide, 1-(2-methylcyclohexyl)-3-phenylurea (siduron), was found to be effective for selective annual weed-grass control in new seedings of forage grasses. In Maryland and Missouri, siduron showed excellent promise for the control of annual weed grasses in plantings of Poa, Festuca, Agrostis, some Bromus species, and many others. Roots of susceptible grasses do not penetrate nor develop in soils containing siduron. Rates ranging from 3 to 6 lb/A were adequate for the control of the weed grasses that germinated on or near the soil surface and rates up to 12 lb/A were tolerated without injury to the other grasses. In Indiana and Maryland, species with larger seeds such as barnyardgrass, giant foxtail, and other species capable of emerging from depths greater than 1 inch escaped injury when this herbicide was applied only on the soil surface. In Washington, siduron controlled 100% of the downy brome and medusahead at rates of 4 to 14 lb/A without injury to the crested wheat-grass planted in September just before preemergence herbicide treatments.

In Maryland, trifluralin applied at 2 lb/A before planting and incorporated to a depth of 5 inches, gave season long control of annual weed grasses. New seedings of alfalfa, birdsfoot trefoil, and crown vetch grew normally in comparison with the growth on untreated and mowed plots in the droughty 1964 season. It was necessary to apply 2,4-DB for control of quickweed, (Galinsoga parviflora), as trifluralin was relatively poor for dicotyledonous weed control. Also, trifluralin, when incorporated in the soil in New York, controlled annual grasses and compared favorably with EPTC at 2 and 4 lb/A when managed similarly. Incorporating EPTC in narrow bands over the seeded row continued to look especially promising for birdsfoot trefoil establishment.

November applications of 1 lb/A 2,4-DB ester and dormant applications of 1/2 lb/A of bromacil eliminated yellow rocket without injury to alfalfa in Missouri.

Excellent weed control for the growing season resulted in plots of big bluestem, Indian grass and switchgrass, sand bluestem and sideoats grama from using atrazine and diuron at 4 lb/A and bromacil at 2 lb/A in Nebraska. There was slight injury to the perennial grasses treated with bromacil early in the growing season, but this was apparently outgrown during the later part of the season.

Stand counts in 1964 showed that repeated sprayings in 1963 with 4 lb/A of isocil and 5 and 10 lb/A of paraquat almost eliminated broomsedge in Mississippi. In other plots, broomsedge fertilized with nitrogen had almost 50% higher protein content in the young stages than those receiving no nitrogen. Six weeks later, most of the difference had disappeared and protein content was much less for all treatments. Nitrogen increased forage yields of broomsedge pastures.

9. Weed Control on Rangelands. A nontillage method of seeding perennial grasses on downy brome-infested rangelands was evaluated for the third year in California and Nevada. In the 1964 experiment, good to excellent initial control of downy brome resulted from spraying paraquat at 0.5 lb/A. First year seedlings of crested and intermediate wheatgrasses drilled in paraquat-treated strips were comparable to those on disked and furrowed plots and about four times better than those on check plots. Results indicate success of this technique.

In Idaho, about 90% control of bracken fern resulted from 2 lb/A of picloram, 8 lb/A of dicamba, and a mixture of dicamba and 2-(2,4-dichlorophenoxy)propionic acid (dichlorprop) at 6 + 2 lb/A. Also, in Washington, picloram at 2 lb/A gave almost complete kill, or inhibition of new rosette formation, of rush skeletonweed while the phenoxy herbicides 2,4-D, 2-methyl-4-chlorophenoxyacetic acid (MCPA) and dichlorprop at rates up to 4 lb/A gave only top kill and appeared to stimulate new rosette formation the fall following application. While Dalmatian toadflax was satisfactorily controlled at 3 lb/A of silvex or dichlorprop, equivalent control was obtained with 1/2 lb/A of picloram. Picloram at 1 1/2 lb/A was required for complete kill.

In Utah, spraying with the phenoxy herbicides just before the bud stage of growth effectively controlled false hellebore. It is probable that one retreatment will be necessary for eradication. Properly timed and repeated annual treatments for 2 or 3 years with 2,4,5-T or silvex was also required for adequate control of tall larkspur from high mountain ranges. Single applications of herbicides did not kill many of these poisonous weeds. The tremendous increase in the grasses and the large reduction of tall larkspur should be sufficient to reduce or prevent cattle losses from poisoning.

On the desert ranges of Utah, halogeton was completely controlled with 1/2 lb/A of paraquat. This herbicide is nonselective but might find use on small infestations or on areas where reseeding can be done immediately following treatment.

In Indiana, it was found that potassium azide was an extremely active herbicidal material whether applied pre- or postemergence. Early evidence suggests that it can kill dormant weed seed in the soil and that it has a short persistence. It also has killed perennial root stocks of Canada thistle.



In Missouri, 2 annual applications of the esters of 2,4-D and 2,4-DB were about equal for controlling ironweed. Surfactants added to 2,4-D and 2,4-DB did not enhance the effectiveness of the esters, but they increased the effectiveness of an amine formulation of 2,4-DB. An application of 1/4 lb/A or more of picloram reduced ironweed stands about 80%. However, it was ineffective on coralberry and wild garlic.

## B. Brush Control

### 1. Physiological and Ecological Studies

Studies in Texas have shown that the herbicidal effectiveness of paraquat and diquat are reduced when spray applications are made at cool temperatures (40-50°F). Laboratory experiments show that light and oxygen are essential for a rapid bleaching of the plant pigment system by paraquat. Light, but not oxygen, is also essential for the changes in membrane permeability brought about by paraquat. Temperature influences changes in membrane permeability. Also, in laboratory studies, picloram inhibited the action of malic dehydrogenase from mesquite. The inhibition was greater than that of 2,4-D and was antagonistic with the oxidized and reduced forms of nicotinamide-adenine dinucleotide. Also a method to quantitatively measure movement of small amounts of 2,4,5-T in plants was developed.

One-seed juniper grows in areas which receive half or more of the annual rainfall during May-October. Utah juniper grows in areas which receive half or less of the annual rainfall during these months. Alligator juniper does not seem to be affected by seasonal precipitation patterns in the Southwest. This information may aid in planning management practices, such as reseeding, following juniper control.

In Texas, it was found that the active translocation area in mesquite stems is a ring about 3 mm thick which includes the outermost xylem ring and the innermost 0.3 mm of phloem. Leaves of plants of one-year-old mesquite, winged elm, and live oak under drought stress in a nursery translocated C<sup>14</sup>-labeled urea, 2,4-D, dicamba and 2,4,5-T poorly when applied in August; but when applied in September to plants that had resumed growth following rain, they translocated these herbicides readily.

The relative importance of stomata, cuticle and trichomes as routes of foliar penetration, was studied by fluorescence microscopy in Arizona. The route of greatest entry is principally dependent upon the plant species. In those species where entry is gained by a combination of two or all three routes, it appears that the nature of the fluorochrome carrier or content of surfactant, may influence one route of entry to the partial or complete exclusion of effect on other routes. For example, some surfactants significantly enhance stomatal penetration, but seem to have relatively little effect on absorption of fluorochromes via trichomes.

Also in exploratory work in Arizona involving the in vitro inactivation of amitrole by mesquite, the relationship between tissue age and the inactivation potential reported last year was corroborated. It has now been observed that the seed is moderately capable of inactivation; but by the age of 4 days, the cotyledons developed an extremely high inactivation potential. By the age of 14 days, this potential is largely lost even though the cotyledons are still green and apparently vigorous.

Absorption and translocation of an ester of 2,4,5-T in mesquite seedlings were significantly enhanced in Arizona by the addition of 40% or more dimethyl sulfoxide (DMSO) to the aqueous carrier. The enhanced activity of 2,4,5-T induced by the higher DMSO concentrations was observed at light intensities ranging from 25 to 1,000 foot-candles. Synergism also existed between DMSO and dicamba or picloram, but to a considerably lesser degree. At a concentration of 20%, DMSO was antagonistic toward the action of all three herbicides. Specific physiological responses to these compounds were generally not related to the light intensity, with the exception that degree of root repression was positively correlated with light intensity. Anatomical and morphological responses to the 2,4,5-T-DMSO mixtures suggest that the latter compound increases uptake and general activity of 2,4,5-T, but that the two substances are probably not transported by the same mechanism or in the same tissues. Whereas, 2,4,5-T translocates principally to the meristematic regions such as the apical bud, the DMSO moves widely throughout the plant and concentrates in the leaf tips. For example, leaves immediately above the treated leaves show marked injury; whereas, such leaves may completely escape visible injury when 2,4,5-T is used alone.

In Texas, a barley huller was modified into an effective mesquite seed thresher. The modified device is capable of threshing one bushel of mesquite pods in 1-1/2 hours. Approximately 150 hours were required to thresh this quantity of seed by hand. Seed of mesquite are required to produce plants for greenhouse and nursery research studies.

## 2. Control Studies

In Nevada, control of green rabbitbrush ranged from 88 to 100% when sprayed with picloram at 1/2 and 1 lb/A, depending on the time of spraying. A June 22nd spraying date gave complete control at both rates while spraying on May 15 controlled green rabbitbrush 88 to 97%, respectively. At these rates of picloram, big sagebrush control was relatively ineffective. 2,4-D at 1 to 3 lb/A in these same experiments gave adequate control of big sagebrush, but control of rabbitbrush was less effective.

Picloram effectively controlled junipers and shrub live oak when applied either as a foliage spray or as soil treatments. Picloram, fenuron, and polychlorobenzoic acid (PBA) all killed most of the grass in the soil-treated area under individual alligator junipers. A marked increase in moss cover occurred under trees sprayed with picloram. Fenuron spot and strip treatments applied in 1961 and 1962 still showed effects on love grasses growing on sandy soil in 1964. Initial responses to large (1/4 inch diameter) pellets of fenuron were equal to or better than standard size pellets, and pellets containing 50% fenuron were equal to or better than 25% pellets applied to a shrub live oak at two different areas in 1964.

In Missouri, picloram applied in 1963 at 1 lb/100 gal. was not as effective as 4 lb/100 gal. for defoliating persimmon in 1964. Soil applications of 5 to 10 lb/A of picloram were about equal to 8 lb/A of dicamba for defoliating persimmon. However, foliage applications of dicamba gave greater control of persimmon than did picloram. Conversely, picloram was more effective on sassafras. In Maryland, picloram controlled multiflora rose growing in hedges whether applied as foliage sprays or soil treatments at the bases of the plant. Only one application, either as a 1 lb/100 gal. of picloram in water spray or 5 lb/A soil treatment was needed in May 1963 to kill existing plants as determined in October of 1964. Native forbs and grasses grew well vegetatively and matured normal seed in 1964.

Picloram at 1/2, 1 and 2 lb/A effectively controlled huisache (Acacia farnesiana). Picloram and bromacil also show promise for control of live oak. Also, paraquat and paraquat combined with other herbicides usually gave most rapid leaf kill of live oak, mesquite, and huisache. It appeared that mixtures of paraquat and picloram retained the rapid action of paraquat and appeared to retain the persistency of picloram on eastern Texas woody plants. In this area, picloram was effective against many woody plant species including sumac, sassafras, chinquapin, and various grapes. In central Texas, picloram was effective for controlling whitebrush when applied at 4 lb/A, which rate also left a good grass stand. Granular applications to the soil were about equally effective as sprays.

Also in Texas, over 97% of the amine salts of 2,4-D, 2,4,5-T and dicamba disappeared within 16 weeks after application to silver and little bluestems and Dallisgrass at College Station, but only 90% of these compounds disappeared in 16 weeks after application to sideoats grama at Spur. Lower rainfall (2.49 inches) was felt to be the main factor accounting for the lower disappearance rate at Spur.



In Puerto Rico, picloram had the broadest spectrum of effectiveness (number of species affected), of the herbicides evaluated. Where defoliation of all species is desired, other herbicides may be required in combination because of differential susceptibility. It was found that defoliation from soil-applied herbicides took place rather slowly but the long-term effects were satisfactory. Soil-applied herbicides were more effective at the site where rainfall was lowest. Results may be influenced either by the amount of rainfall or by completely different species composition on the different sites. There was greater species diversity found at the Maricao site where the rainfall was intermediate and soils more porous than at the relatively wetter Luquillo site or at the drier Guanica site. Better soil drainage at Maricao prohibits soil saturation for an extended period.

Paraquat and diquat will defoliate guava within a week in Puerto Rico, but recovery was complete at the end of 6 months. Conversely, picloram was most effective in killing guava, but the action was somewhat slower than that of paraquat and diquat.

In both Oklahoma and Texas, picloram shows promise for control of winged elm which has been tolerant of 2,4,5-T sprays. In both Oklahoma and Missouri, picloram was ineffective for the control of Symphoricarpos species. When applied as pellets to the soil around each cactus plant, picloram was effective for its control in Oklahoma. Also in Oklahoma, it was found that only small oak brush (1-3 inches in diameter) was killed by two annual April burnings. Trees larger than 3 inches in diameter were not affected. There was considerable more sprout growth on areas burned than on the untreated. On the other hand, 2,4,5-T ester applied either as a foliar spray or by injector gave good control of both small and large post and blackjack oaks. Native grass release and growth were best on 2,4,5-T treated plots. There was a significantly higher moisture content in the soil throughout the growing season through the 4 feet of sampling depth on 2,4,5-T treated plots as compared to those burned. Soil moisture was about the same for burned as for untreated.

In Oklahoma, triiodobenzoic acid added at 1/16 and 1/8 lb/A with 2 lbs. of an ester of 2,4,5-T per acre as foliage sprays gave nearly twice as much control of post and blackjack oak as spraying with an ester of 2,4,5-T alone. Oak defoliation was also more with the addition of 1/80 and 1/40 lb/A of thiocyanate to an ester of 2,4,5-T.

### C. Nematode Control

1. Forage and Range. Cooperative investigations with Auburn University indicated that a new vetch variety 'Warrior' (Vicia sativa) was highly resistant to root-knot nematodes (M. incognita incognita, M. incognita acrita, and M. javanica). No resistance could be found in vetches to M. arenaria and M. hapla. Similarly, several Sericea lespedeza breeding lines were highly resistant to one or more root-knot species (M. incognita, M. incognita acrita, and M. hapla); however, these lines were highly susceptible to M. arenaria and M. javanica. Studies on the nature of resistance of alfalfa to the root-knot nematode (M. hapla) at Logan, Utah indicate that resistant lines, as determined in greenhouse tests, have no more resistance than nonresistant selections when foliage is clipped. Studies are underway to determine the nature of resistance of alfalfa to the stem nematode (Ditylenchus dipsaci) for areas where 'Lahontan' cannot be grown; 'Kayseri' shows 67% as much resistance as that of 'Lahontan'. Environmental studies have indicated that approximately 10 days of warm, dry weather during the middle of May reduces field infestations of the stem nematode in northern Utah. The effect of the environment on the occurrence of this nematode will be studied in more detail. Varieties and strains of bermudagrass were studied for susceptibility and population increase of sting nematode (B. longicaudatus) at Tifton, Georgia. Arizona common and coastal bermudagrass were susceptible to the sting nematode but good resistance is available in a cross of coastal bermuda x PI 255445 F, 29-6. None of the bermuda lines showed appreciable resistance to the stubby-root nematodes. Coastal bermuda supported relatively large populations of sting, stubby-root, and ring nematodes but effectively suppressed population development of the root-lesion nematodes. Populations were lowest for all nematodes on the cross of coastal bermuda x PI 255445 with resistance apparently coming from the PI 255445 parent.

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CORN, SORGHUM, AND SMALL GRAIN INSECTS  
Entomology Research Division, ARS

Problem. Many species of insects cause losses amounting to millions of dollars annually to corn, sorghum, and small grains. It is estimated that 25 species of insects cause an annual loss of \$900 million to corn alone. The European corn borer and corn earworm are two of the most destructive insects in the country, and corn rootworms are serious pests of corn. Armyworms attack corn and small grains. In certain years the greenbug causes widespread losses to wheat, barley, and oats in the Central and Southeastern States, and the Hessian fly and wheat stem sawfly annually damage the wheat crop in certain areas. The cereal leaf beetle, first identified in the United States in 1962 from Berrien County, Mich., now occurs in 68 counties in Michigan, Indiana, and Ohio, and is a threat of unknown proportion to small grain crops. Such examples of the destructiveness of insects to corn, sorghum, and small grains point up the need for extensive research that will lead to the development of adequate means for the control of these important crop pests. Progress has been made toward the solution of some of the insect problems encountered in the production of grain crops but more effective, more economical, and safer insect control measures are needed. Research is essential to find insecticides that can be applied to grain crops, that will not leave residues harmful to animals consuming the feed, that will not be a hazard in milk and meat, and that will not be detrimental to beneficial insects or to fish and wildlife. The appearance of resistance to certain insecticides in several grain insect pests stresses the need for basic information to overcome this problem. Additional emphasis should be placed on research to develop crop varieties resistant to insects and on biological and cultural control methods. New approaches to insect control, such as sterilization techniques and attractants, require expanded investigation. Research is also needed on insect vectors and the role they play in the dissemination of important plant diseases. The heavy losses in oats, wheat, and barley due to barley yellow dwarf virus, and in corn due to maize dwarf mosaic and corn stunt recently found in several North Central and Southern States, indicate the importance of research in this field.

USDA AND COOPERATIVE PROGRAM

The Department's program involves both basic and applied research directed toward developing more efficient control methods for insects attacking grain. All studies are conducted in cooperation with State Experiment Stations in the several States where research is underway. Studies on evaluating and developing varieties of grain which resist insect attack are conducted in cooperation with State and Federal agronomists and plant breeders and research on insect transmission of diseases of grain crops is in cooperation with State and Federal plant pathologists. This research includes studies on Hessian fly, wheat jointworm at Lafayette, Ind., and Manhattan, Kans.; cereal leaf beetle at Lafayette, Ind., and East Lansing, Mich.; aphids and mites attacking small grains at Stillwater, Okla., Brookings, S. Dak., and Tifton, Ga.; wheat stem



sawfly at Fargo, N. Dak., and Bozeman, Mont.; corn earworm at Tifton, Ga., State College, Miss., and Lafayette, Ind.; fall armyworm, pink scavenger caterpillar, and rice weevil at State College, Miss., and Tifton, Ga.; soil insects attacking corn at Brookings, S. Dak., State College, Miss., and Tifton, Ga.; corn leaf aphid at Brookings, S. Dak.; southwestern corn borer at Stillwater, Okla., and State College, Miss.; European corn borer at Ankeny, Iowa, State College, Miss., and Wooster, Ohio; corn earworm, sorghum midge, sorghum webworm, and corn leaf aphid on sorghums at Stillwater, Okla., and Tifton, Ga.; and insect transmission of grain diseases at Manhattan, Kans., State College, Miss., and Brookings, S. Dak. Research to evaluate improved equipment for application of insecticides to grain crops is underway at Ankeny, Iowa, and Tifton, Ga., in cooperation with Federal agricultural engineers. Work on corn rootworms is being conducted at Brookings, S. Dak. Additional research is being conducted under ARS contracts and grants on the biology and control of the cereal leaf beetle with Michigan, Indiana, and Ohio Experiment Stations, soil insects attacking corn with the University of Nebraska, and vectors of corn stunt virus with Mississippi State College, nature of resistance of corn to the European corn borer with Iowa State University, and insect communication in the infrared region with Michigan University, Ann Arbor, Mich.

The Federal scientific effort devoted to research in this area totals 42.5 professional man-years. Of this number 10.8 is devoted to basic biology, physiology, and nutrition; 3.5 to insecticidal and cultural control; 1.8 to insecticide residue determinations; 4.1 to biological control; 2.9 to insect sterility, attractants and other new approaches to control; .5 to evaluation of equipment for insect detection and control; 15.7 to varietal evaluation for insect resistance; 1.8 to insect vectors of diseases; and 1.4 to program leadership.

Certain phases of this research are contributing to regional research project NC-20 "Factors Influencing European Corn Borer Populations". A P. L. 480 project, E8-ENT-1, "Population Dynamic Studies on Calligypona pellucida (F.) and the Nature of Injuries Caused by This and Other Leafhopper Species (Fulgoridae) on Cereals, Especially Oats and Spring Wheat" is underway at the Agricultural Research Centre, Department of Pest Investigation, Helsinki, Finland. Another P. L. 480 project, A10-ENT-5, "Host Plant-Vector and Host Plant-Virus Relationships of Rough Dwarf Virus of Corn and Methods for Control of the Disease" is being conducted at the Hebrew University, Rehovoth, Israel. A7-ENT-25 in India is concerned with "Research on Insect Pests of Maize With Special Reference to Stalk Borers".

#### PROGRAM OF STATE EXPERIMENT STATIONS

Extensive research is in progress in the States on insects affecting corn, sorghum and small grains. Biological information is being obtained on a variety of pests. Data acquired include overwintering habits, time of emergence, food habits, interspecies competition, mating, oviposition, migratory, and dispersion habits and longevity. This information is being



used to develop methods of predicting the incidence of pest outbreaks.

Ecological studies are being performed to determine the effects of temperature and other factors such as plant growth, soil conditions, and crop sequence on population levels. The influence and efficiency of various natural enemies are also being evaluated.

Cultural control techniques including the effects of fertilizer applications, soil management practices, time of seeding, irrigation, stubble mulch, and grazing receive their share of attention.

Experimental insecticides are tested for their effectiveness. Samples of treated crops are analyzed for harmful residues. Insecticide treatments are also used to determine the degree of infestation which crops can tolerate before control becomes necessary.

Research on artificial rearing is performed to develop methods for providing insects for year round study and for uniform infestation in plant resistance work. Plant varieties, hybrids and lines are evaluated in the field and in nursery plots for their resistance to insect attack. Crosses are made to increase resistance levels and biological, physiological, and chemical studies are conducted to determine the nature of the resistance.

Vectors of plant diseases are studied to increase our knowledge of insect-plant relationships which could lead to the control of the vector and, consequently, the disease. The roles of alternate host plants and vector seasonal life history and flight patterns are being studied. The effect of the disease organism on vector biology, morphology, and cytology is investigated.

There are 49.5 professional man-years devoted to research on corn, sorghum, and small grain insects in the States.

#### PROGRESS USDA AND COOPERATIVE PROGRAM

##### A. Basic Biology, Physiology and Nutrition

1. Corn Insects. Studies were continued on interrelation of factors affecting control of the European corn borer in Boone County, Iowa. The 1964 early spring survey showed a population of 4,228 borers per acre, and the late spring survey showed 3,250 borers per acre. This was a reduction of 64.2% from the postharvest survey conducted in 1963. The midsummer borer population in 1964 averaged 5,416 borers per acre and 94% of the first-brood borers pupated. The 1964 fall population averaged 7,478 borers per acre prior to harvesting whereas the postharvest survey revealed 2,040 borers per acre.

Light trap catches at the Ankeny farm indicated only 2 generations of corn borer were present during the 1964 growing season.

In connection with NC-20, laboratory and field experiments were conducted with borers from Minnesota, Iowa, and Missouri to investigate the possibility of the existence of biotypes. The results indicate that there are at least 2 biotypes of the corn borer, the northern in which a percentage of the population undergoes obligatory diapause and the southern in which a large percentage of the population undergoes facultative diapause. It appears that diapause in the corn borer is determined genetically in addition to other factors that have been demonstrated to cause this condition. The results of the experiments lead to the theory that diapause in this insect is governed by a multigenetic makeup which responds to changes in temperature and photoperiod. The concentration of the genes in an individual determines the extremes of temperature and photoperiod that can be experienced before diapause is induced. Thus, an individual larva with a low concentration of diapause inducing genes could withstand shorter days and cooler temperatures without inducing diapause than one with a high concentration of diapause inducing genes.

Methods of rearing European corn borer in the laboratory have been further refined. When strips of corrugated paper were placed in the rearing container mature larvae entered the paper to pupate. The paper can then be transferred to emergence cages without handling each pupa. Leaf factor material used in the diet is now cut with a field chopper and dried in a commercial grain dryer.

The laboratory colony of borers begun in August of 1963 has been continued and is now in its 25th generation. No major change in the insect is evident from the early generations to the 24th, although there may be a trend for pupal weights to be slightly higher. This colony, and 3 sub-colonies, have provided egg masses for year round use in all laboratory studies of the various projects.

A study at Wooster, Ohio, indicates that the chromosomes of the European corn borer have a diffuse centromere, thus broken chromosomes would migrate to the poles at meiosis just like whole chromosomes.

In studies at Brookings, S. D., it was found that larvae of the western corn rootworm required a relative humidity of 98% or above to survive more than 11 hours. Therefore, larvae hatching in the top few centimeters of dry soil exposed to direct sun might be desiccated before finding food and a suitable environment.

Western corn rootworm eggs exposed to temperatures of 5° F or below for one week did not hatch while eggs exposed to 14° F hatched. Even in severe winters in South Dakota temperatures in the soil where the eggs are seldom go below 14° F.

The recent outbreaks of resistant western corn rootworms and spread of these resistant populations to new areas of the Corn Belt have brought this insect and the northern corn rootworm into overlapping areas, and the two species have



been observed mating. In laboratory matings of the two species, all offspring reared to the adult stage were, by all available criteria, western corn rootworms. The few individuals reared to the adult stage were inbred and one male was backcrossed to a lab reared western female. The hybrid laid 329 eggs, 90% of which hatched. The backcross was also fertile, which ruled out male hybrid sterility. Further inbreeding through three generations produced fertile offspring resembling the western corn rootworm.

Larvae of the western corn rootworm have been reared to adults on a diet consisting of a variation of the pink bollworm diet with an additional corn kernel plant factor, but the development was slow and small adults with deformed wings were produced, indicating nutritional deficiencies and possible absence of feeding arrestants.

The feeding, resting, and movement behavior of the northern, western, and southern corn rootworms were compared under field conditions. All three species preferred succulent silks and ears for food when present. The northern corn rootworm was found in larger proportions than the western corn rootworm on these parts. Greatest foliage feeding by the western corn rootworm was apparent. Adult migration and movement was strongly correlated with increased corn maturity and suggested that movement or spread of the western species to new areas occurs during the late season period when older corn is less desirable as food. The southern corn rootworm moves to other crops after adult emergence in corn fields. The percentage of the aldrin and heptachlor resistant western corn rootworms to total rootworm population gradually increased from practically none in 1962 to 22% in 1964 in the Brookings, S. D., area.

In Nutritional studies with the corn leaf aphid, one diet was developed which could sustain survival and reproduction over a three week period. A 20% sucrose solution was optimum in basic diets. Water extracts from seedling corn incorporated in the diets failed to inhibit aphid survival or reproduction. It was concluded that young plants contain no deleterious substance to this aphid species even though it does not colonize on seedling corn under field conditions.

A mass rearing method for the southern and western corn rootworms has been developed at Brookings, S. D. The relatively simple procedure involves larval development in shallow screen-bottom trays thickly planted with corn in a 1:1 vermiculite-soil mixture. The dense root mat in a single 12" x 18" tray supplies ample food to mature 200-250 larvae. A single transfer of the infested mat to a greenhouse flat affords an ideal site for pupation and adult emergence.

The corn earworm continues to be the most important corn insect in the South and caused an estimated loss of over \$5,000,000 in the 6-State area of Alabama, Florida, Georgia, North and South Carolina, and Mississippi in 1964. The southwestern corn borer has continued to spread in Mississippi and Alabama.



At Tifton, Ga., a vitamin mixture, recommended and commonly used for rearing lepidopterous larvae, was found to be deficient in two vitamins, folic acid and vitamin B<sub>12</sub>, when used in a fall armyworm diet. The substitution of another vitamin mixture containing the two vitamins in sufficient amounts has almost entirely eliminated insect deformities experienced in previous attempts to rear this insect.

Research on the theory that insects locate and communicate by means of infrared and microwave radiation has continued at Tifton, Ga. Experiments with flight activity of noctuid moths under different lighting conditions have shown that mating is highest with ultraviolet and cool-daylight fluorescent lights alternating with darkness. The results indicate that conditioning the insects with ultraviolet and visible radiation contribute to their efficiency of receiving infrared and microwave radiation in darkness.

An infrared blackbody was constructed which eliminated all visible light but allowed radiation of secondary emission in the 8 $\mu$  to 13 $\mu$  region. Six different species of noctuid moths were attracted to the radiating blackbody in a totally dark room, whereas nonemitting blackbody controls did not attract any moths.

The following percentages of infrared transmission were obtained when the corneal lens of sphingid moths were cleaned and mounted in an infrared spectrophotometer: 2.5 to 2.85 $\mu$ , 40%; 3.5 to 5.7 $\mu$ , 50%; 7.5 to 8.8 $\mu$ , 15%; and 10.3 to 13.0 $\mu$ , 40%. All fall within atmospheric windows, indicating that the outer lens of the moth eye can transmit infrared radiation.

In Mississippi biological and ecological studies of the southwestern corn borer revealed that moths were attracted to mercury vapor and black light traps set up adjacent to corn fields. Mercury vapor light was more attractive than black light. Biological investigations showed that diapause was induced by 13 and 14 hours of light, although twice as many larvae entered diapause at 13 hours than did at 14 hours. In light chambers used for this study diapause was prohibited by using a 30-minute light period daily in the middle of the 11-hour period of darkness.

Research conducted in India under P. L. 480 project A7-ENT-25 showed that Chilo zonellus is the major pest of maize and progress was made on mass rearing of this pest and also Sesomia infirens.

2. Small Grain and Sorghum Insects. At Brookings, S. D., successive generations of a false wireworm Eleodes suturalis have been continuously colonized and mass produced in the laboratory. The continuous availability of a coleopterous soil inhabiting insect provides a test organism for soil insecticide screening and bioassay, as well as a subterranean species for basic research in fields of toxicology, physiology, and ecology.

At Tifton, Ga., the corn earworm and fall armyworm utilization of sorghum leaves and heads was compared with corn leaves and kernels. Although smaller

in size, the fall armyworm larvae utilized more of each plant part than did the corn earworm larvae. Both species of insects utilized corn kernels better than corn leaves, sorghum heads, or sorghum leaves. Corn plant parts were utilized to a greater extent by both species than sorghum plant parts.

Environmental factors responsible for corn leaf aphid flight initiation have been studied at Tifton, Ga. There was a significant positive correlation between actual barometric pressure and number of aphids initiating hourly flight, and there was an indication that high concentrations of negative ions increased aphid flight.

At Stillwater, Okla., artificial diets for rearing lepidopterous larvae have been improved by increasing the amount of formalin and adding propionic acid. This reduced mold development, and larval production was increased.

Third instar corn earworm and fall armyworm larvae reared on artificial diets, when placed on 30-day old RS 610 sorghum in the greenhouse, fed readily, and the leaves showed damage typical of naturally infested sorghums. This test demonstrated the practical use of larvae mass-reared on artificial diets for use in manual infestations in connection with searching for resistant germ plasm.

Work was initiated at East Lansing, Mich., on mass rearing of the cereal leaf beetle. Field collected and laboratory reared adults were capable of mating and egg production after 6 weeks' hibernation when confined in an oviposition chamber at 40° F. After hatching the larvae are transferred to flats of barley, where the soil of the flat has been covered with plaster of paris and sand, to complete larval development and pupation. Pupae are removed and held for adult emergence. This method of rearing has provided access to the pupal stage and has greatly increased production of adult forms.

Flight of the cereal leaf beetle was found to be greater at temperatures above 75° F than at lower temperatures; between 12:00 noon and 3:00 p.m. than at other times; at 3 feet than at 6 or 11 feet; and with summer adults than with spring adults. Temperature and the related time of day proved to be the most important factors affecting flight. The pest tended to fly with the wind, but the influence of wind velocity and cloud cover were minimal on the amount and height of flight. Large numbers of both spring and summer adults were found to move 1-1/2 miles during their period of activity.

Developmental limits of the cereal leaf beetle were found to be 52° to 90° F with an optimum of 80° and 85° F for all stages. The diapausing adult was able to survive at 0° F and all stages could survive at 110° F. Three years of study of the pest in the field have shown that the different stages were found to be "common" (i.e. easily found) at: spring adults, 37.5 to 68.5 day-degrees with a base of 52° F; eggs, 65.5 to 90.8 day-degrees; larvae, 249.3 to 250.5 day-degrees; pupae, 510.5 day-degrees; summer adults, 764.5 to 786.5 day-degrees.

A 1:1 sex ratio was determined for both spring and summer adults of the cereal leaf beetle and egg production is estimated at 96 eggs per female in the field. Natural mortality of the different stages of the pest is estimated at: eggs, 34% in winter wheat, 19% in spring oats; larvae (the greater part in failure to enter pupation) 68% in wheat, 60% in oats; pupae, 68% in wheat, 60% in oats; 60% of the overwintering adults; and minor mortality of active spring and summer adults. The differences in egg mortality between wheat and oats is caused by lower temperatures during the period that the eggs are in the wheat than in the oats and in almost equal part by predation, principally by the lady beetle Coleomegilla maculata lengi in the wheat. Mortality during the pupal stage (including failure of the larvae to form pupae) is attributed to high soil temperatures and the overwintering kill is attributed to prolonged exposure to low temperatures with moisture and predation being of relatively minor importance. With about 3.4 times more eggs layed in oats than in wheat, the population increase of the pest is estimated at 369% per year.

## B. Insecticidal and Cultural Control

1. Corn Insects. Twenty-four insecticides were tested in granular formulations against first-generation European corn borer larvae at Ankeny, Iowa. The most effective compound in the test was methyl parathion at 1.0 pound per acre but this was not significantly better than DDT at 1.0 pound per acre. Thirteen other compounds gave good control. Of the 16 compounds tested in granular formulations for control of second-generation borers, 10 appeared to be as effective as DDT. Fifteen insecticides were tested in spray formulations against first-generation corn borer and 7 were more effective than DDT.

Of several compounds tested for systemic control of corn borer larvae in field corn, only American Cyanamid 47470 at 2.0 pounds and 4.0 pounds actual insecticide per acre was effective.

In toxicological studies with the western corn rootworm at Brookings, S. D., the aldrin LD<sub>50</sub> values for third instar larvae were higher than for adults of aldrin resistant western corn rootworm collected from the same fields. There was a definite trend toward decreasing LD<sub>50</sub> values for adults as the growing season progressed. The average aldrin resistance in the eastern range of distribution of western corn rootworm has increased 26 to 199% over the levels obtained in the same localities in 1963. Although there are scattered areas where northern corn rootworm manifest aldrin resistance, the incidence of resistance is relatively static. Both species of corn rootworm appear to remain quite susceptible to the action of diazinon and phorate.

Previous studies indicated that the resistance mechanism in western corn rootworm involved the epoxidation metabolism of Cl<sub>36</sub> labelled aldrin and dieldrin. Preliminary studies with C<sub>14</sub> labelled material indicate that there were at least two additional major metabolites (besides aldrin and dieldrin) not present in the Cl<sub>36</sub> studies. It is possible that one of these metabolites represents the dechlorinated insecticide.



Varied cultural practices were investigated for western corn rootworm control. Different discing and plowing sequences in both the spring and fall had no significant effect in reducing rootworm larval populations. There were 7 times more overwintering rootworm eggs in plots where corn was allowed to mature than when cut for silage in late August.

At Tifton, Ga., five applications of any of the following compounds when applied at the rate of 1 pound per acre gave corn earworm control equal to or better than 2 pounds of DDT per acre: SD-9129 and SD-8447, Mobil Oil MCA-600, Niagara NIA-10242, UpJohn U-12927, Stauffer R-5092, and General Chemical GC-4072.

In Mississippi early planting was found to minimize southwestern corn borer injury to corn. Three weekly applications of endrin at 0.5 pound per acre starting when second generation larvae hatched, gave 77.2% control.

Research conducted in Nebraska under contract showed that the western corn rootworm was developing resistance to phorate in some areas. Several experimental organic phosphate insecticides appear to be as effective or better than those now recommended.

2. Small Grain and Sorghum Insects. At Stillwater, Okla., hybrids and parental lines of sorghum were sprayed with insecticides at recommended rates to measure differential insecticide phytotoxicity. All entries except RS 610 and RS 626 showed phytotoxicity following spraying with methyl parathion and Bidrin, with leaf area damage ranging around 50%. Ethion, diazinon, and carbaryl W.P. and the water sprayed check plots did not show injury. Yield reductions resulting from phytotoxicity were not as great in 1964 as they were in 1963. Temperature and plant condition, undoubtedly, are associated with insecticide phytotoxicity and yield reduction.

Several insecticides were found to be superior to parathion for greenbug control in laboratory tests. These included emulsifiable concentrates of GC-6506 and GC-3707. Two 10% granular formulations, UC-21149 and NIA-10242, provided a faster "knockdown" than Di-Syston granules and provided 100% greenbug control at the rate of 0.063 pound per acre under greenhouse conditions.

In greenhouse insecticide screening tests there were indications that greenbugs might be developing resistance to parathion. Previously, 100% control was obtained in the laboratory with parathion spray at the rate of 0.016 pound per acre, but more recently, the rate of 0.125 pound per acre (8 times the rate used formerly), failed to give satisfactory control.

In Montana no insecticides tested for control of wheat stem sawfly were as effective as heptachlor. An additive, dimethyl sulfoxide, appeared to enhance the penetration of Meta-Systox when applied to wheat foliage.

In aerial application tests conducted under contract in Michigan, technical malathion applied at the rate of 4 liquid ounces per acre was as effective as 5 liquid ounces against the spring adults of the cereal leaf beetle. The use of "mini-spin" nozzles for the aerial spray resulted in a slower initial kill but a longer residual effectiveness of the malathion than did flat tip nozzles.

Laboratory tests were conducted to determine the toxicity of carbaryl, malathion, and dieldrin to cereal leaf beetle adults. Topically applied concentrations of the insecticides demonstrated LD<sub>50's</sub> of 0.007%  $\mu$ g per beetle, 0.01  $\mu$ g per beetle, and 0.018  $\mu$ g per beetle for carbaryl, dieldrin, and malathion, respectively.

ENT-25784, endosulfon, Meta-Systox R, Geigy 13905, General Chemical 9160 and 4072, Niagara 10242, Shell 8530, ENT-25736, Baygon, Mobil MCA-600, and Stauffer B-10119 as foliage sprays, and Union Carbide 21149 as a soil-applied systemic, showed promise against the larvae of the cereal leaf beetle.

### C. Insecticide Residue Determinations

1. Residues on Corn. Soils from Ankeny, Iowa, were analyzed for American Cyanamid CL47470. Samples were taken at 2 to 6 inches, 6 to 10 inches, 10 to 14 inches, and 14 to 18 inches at 0, 1, and 7 days after the compound has been applied to the soil in a band parallel to the corn row at a rate of 2 pounds per acre. Results of these analyses indicate that there was very little movement of the pesticide in the soil during the 7-day period.

At Tifton, Ga., in studies of Shell SD-8447 applied to sweet corn as a wettable powder at rates of 1/2, 1, and 2 pounds per acre, electron affinity gas chromatography analysis revealed that initial residues on the stalks and leaves were about the same as those on ear husks. However, the residues on the husks diminished faster. The level of residue on both plant parts varied directly with the quantity of insecticide applied and after 16 days of weathering only 0.42 and 0.02 ppm of the insecticide remained on the stalks and leaves and on the ear husks, respectively, in plots treated with the highest rate. No detectable residues were found in the ears after 16 days weathering in the field.

2. Residues on Small Grain and Sorghum. Heptachlor was applied as a 5% granular formulation in the furrow with wheat seed at planting at the rate of 1/2 or 1 pound per acre. No measurable residues of heptachlor were found in the green wheat 71 days after planting, but residues of heptachlor epoxide were 0.011 ppm for the 1-pound dosage and 0.015 ppm for the 1/2-pound dosage. When the grain was harvested 146 days after planting, no measurable residues were found in the grain. Straw samples taken at harvest contained no measurable residues of heptachlor, but heptachlor epoxide residues of 0.010 ppm for the 1-pound dosage and 0.045 for the 1/2-pound dosage were present.

### D. Biological Control

1. Corn Insects. Field and laboratory tests with the bacterium Bacillus



thuringiensis in Iowa continue to show its potential usefulness as a microbial insecticide for European corn borer control. Granule formulations controlled field infestations of the European corn borer as well as the recommended insecticide DDT. Preliminary field tests with encapsulated bacterium also indicated good borer control. The encapsulation process appears to provide a broad spectrum of opportunity in insecticide and microbial formulation. Capsules can be tailored to size and conditions of breakdown. Both liquids and dry material can be encapsulated. Other forms of life than those to be controlled can be protected from toxic chemicals, tastes can be masked, and microorganisms susceptible to environmental conditions can be protected. Formulations containing materials for immediate release can be applied at the same time with materials encapsulated for later breakdown and release.

Investigations involving the borer-infecting protozoan Perezia pyraustae indicates that the epithelial lining of the young borers' alimentary tract is heavily invaded, but the infection in this tissue is transitory. Under field conditions such larvae would have trouble becoming established, since the gut wall cells could not function normally.

The distribution and abundance of exotic parasites of the European corn borer were determined from larvae collected in 14 States. Parasitism was above 10% in Iowa and Maryland, and ranged from 1.5 to 6.9% in the other 12.

A nematode, Mesodiplogaster sp., isolated from soil at Brookings, S. D., has proved to be lethal to infected corn rootworm larvae and pupae. All infected insects perish in one to three days. The beetle or eggs are apparently not affected. The nematode is easily reared aseptically on cooked pork kidney cubes placed on water agar slants. Preliminary research in Illinois has shown this organism will also infect and kill the onion maggot.

At Tifton, Ga., field releases have been started on two species of tachinids obtained from India in connection with P. L. 480 projects. The species, Drino imberbis and Piparyia tachinid sp. #3, parasitize large corn earworm larvae and are easily reared in the laboratory. They will also parasitize the tobacco budworm and the fall armyworm but appear to develop better on the corn earworm. A small braconid parasite from India, Microplitis sp., was able to parasitize both the corn earworm and the tobacco budworm in the laboratory.

Native parasites were reared from corn earworm collected on wild host plants. Microplitis croceipes was reared from the corn earworm collected on crane's bill. Campoletis sp. was found parasitizing corn earworm larvae on toad flax. Two species of tachinids were reared from fall armyworms collected on Coastal bermudagrass and millet. The more abundant tachinid was Lespesia archippivora. Winthemia rufopicta was less abundant. A colony was established of Lespesia archippivora which proved easy to rear in large numbers using the same techniques which were developed for Drino and Piparyia. An egg parasite, Trichogramma sp., was collected in the corn field early in the summer by exposing corn earworm eggs on pieces of paper napkin clipped to corn leaves.



At Tifton, Ga., the nuclear polyhedrosis virus of the corn earworm was used in an early season field trial on sweet corn in which the virus alone and in combination with DDT was compared with a DDT standard and an untreated control. The best control was obtained with a combination of virus and DDT applied at the early-tassel treatment and at 3-day intervals during silking. There was no significant difference between: (1) DDT alone applied to the tassel stage followed by treatments at 3-day intervals in the silking stage, (2) the virus alone applied to the tassel stage followed by treatments at 3-day intervals during silking, or (3) a single virus treatment applied during the tassel stage. All were significantly better than the untreated control.

2. Small Grain and Sorghum Insects. In Finland studies were continued on population dynamics of the leafhopper, Calligypona pellucida under P. L. 480 project E8-ENT-1. A triungulid Elenchus tenuicornis was found to be an important parasite of C. pellucida and the biology of the parasite has been investigated.

#### E. Insect Sterility, Attractants, and Other New Approaches to Control

1. Corn Insects. Results of investigations carried on during 1964 at Ankeny, Iowa, indicate that chemosterilants can be used to sterilize European corn borer adults by treating partially developed larvae. When 5-day-old larvae were placed for 24 hours on hempa-treated diet the resulting male moths were 90% sterile. The longevity and mating rate of the treated moths compared favorably with the untreated moths. Two to four times as much chemosterilant was required to sterilize the female moths as the male moths.

Third, 4th, and 5th instar corn borer larvae have been irradiated at dosages ranging from 2,000 to 20,000 roentgens, in 500-r increments, to determine the effect of gamma rays on growth, pupation, mating, egg production and hatch. Dosages above 10,000 r prevented larvae from pupating but did not prevent their growth to pupation size. Although pupation was nearly normal at 5000 r, the adults which emerged were abnormal morphologically. At dosages up to 4000 r, growth, pupation, emergence, and mating were apparently normal. From these experiments it would appear that 3rd instar larvae, or larvae in diapause, could be sterilized practically with a dosage of about 3000-3500 r.

In connection with the irradiation experiments, histological and cytological studies are being carried out on spermatogenesis and sperm formation in normal, irradiated, and chemosterilant-treated European corn borers. Testes are fixed, embedded, and sectioned for both light and electron microscopy. These studies indicate that 3rd instar testes contain mostly primary and secondary spermatogonia, 4th instar testes contain all stages up to early spermatids, and 5th instar testes contain even mature spermatozoa. Testes in diapausing 5th instar larvae contain mostly spermatocytes in an arrested stage of development.

At Tifton, Ga., testing of ether extracts from virgin fall armyworm, corn earworm, and armyworm moths has shown that a specific mating stimulant (sex pheromone) is produced by the female of each species. In all three insects the

lure was found within the last two abdominal segments. Attempts to recover the pheromone from heads, thoraces, and/or upper abdominal segments have failed.

When male fall armyworm moths, 3 to 6 days old, were exposed to 0.2 equivalents of females of different ages, only 2% of the males reacted to extracts of females 12 hours old, 32% reacted to extracts of females 24 hours old, 80% to females 36 hours old, and 98% to females 48 hours old. When 3- to 6-day-old males were exposed to varying concentrations of extracts of 3-day-old females, 98% of the males reacted to 0.02 female moth equivalents (FME), 88% reacted to 0.002 FME, 66% reacted to 0.0002 FME, and 20% reacted to 0.00002 FME.

At Tifton, Ga., tepa was found to sterilize the armyworm, *Pseudaletia unipuncta*. Treating fall armyworm moths with hemel and hempa by feeding, tarsal contact, or topical application did not reduce egg hatching, number of eggs laid, or mating frequency. Male fall armyworm moths sterilized with tepa competed with untreated males for untreated females and mated as frequently as did the untreated males.

2. Small Grain and Sorghum Insects. By the use of a specially designed soil olfactometer, a substance has been discovered which attracts larvae of *E. suturalis*. Larvae move through soil in response to the attractant. The substance appears to be a product of seed germination. Air passed over wet or dry wheat kernels or over ground wheat will not attract. Although it appears to be a product of seed germination, there seems to be a concentration factor where it becomes a repellent or a second substance or substances become a repellent at high concentrations.

In studies conducted under contract in Michigan, diapausing females of the cereal leaf beetle were sterilized by exposures of 3000 roentgens of radiation. The gonads of the females do not mature until after they have undergone diapause, but the male gonads are fully developed upon emergence and before diapause.

In Michigan 370 synthetic compounds were evaluated as possible attractants for spring adult cereal leaf beetle and 250 for summer adult beetles. None were highly attractive to the beetle, but a few showed responses greater than the check. All tests with natural lures were negative.

Ether, ethanol or water extracts from barley, oats, wheat, corn, or beans induced no response to cereal leaf beetle adults. However, seedling plants of barley or oats when placed in bean fields attracted many beetles. When 16 different colors were tested for response of cereal leaf beetle, counts of beetles collected indicate a high preference for canary yellow, followed by white, ivory, orange, and pink, in that order. Blue, red, and black attracted very low numbers, black being nearly nonattractive.

#### F. Evaluation of Equipment for Insect Detection and Control

1. Corn Insects. At Tifton, Ga., entomologists in cooperation with



agricultural engineers, installed a fan on an ultraviolet light trap in order to force the "catch" into a treating chamber. The chamber is coated with a chemosterilant suspended in a 10% sugar solution and allowed to dry. Fall armyworm and corn earworm moths come in contact with the chemosterilant by feeding and/or by tarsal contact. After predetermined exposure the moths are allowed to escape back onto the environment for competition with untreated moths for mates.

At Tifton, Ga., agricultural engineers worked with entomologists and chemists to develop a high clearance tractor-mounted revolving brush applicator to treat corn silks for corn earworm control. Applications at the rate of 1.3 pounds per acre produced two to four times more insecticide residue on ear tips than emulsion sprays applied at the rate of two pounds per acre. The brush applications resulted in as good earworm control as that obtained with emulsion sprays.

Agricultural engineers and chemists at Tifton, Ga., evaluated the efficiency of an electrostatic duster to place dust on the upper and lower surfaces of leaves. Utilizing DDT dust and 10-day-old bean plants they found that deposits of insecticide on the top surfaces of leaves were about 57% greater from charged nozzles than from uncharged nozzles and that residues on the bottom surfaces were about 310% greater from charged nozzles. In tests with uncharged nozzles, the quantity of insecticide found in the bottom surfaces of the leaves represented about 11% of the total on both sides, while with the charged nozzles the residue on the bottom surfaces accounted for about 25% of the total.

2. Small Grain and Sorghum Insects. Four types of traps are being used in South Dakota for sampling of aerial cereal aphid populations. These include yellow pan, Shands type directional wind traps, a revolving net trap, and a Johnson-Taylor suction trap. Yellow pans have proven to be ineffective cereal aphid traps in South Dakota but are being continued in cooperation with NC-67. Shands trap catches vary directly with wind velocity and are useful for detection of aphids but not for population comparisons between years or areas. The revolving net and Johnson-Taylor traps are efficient aphid catchers and sample constant volumes of air.

#### G. Varietal Evaluation for Insect Resistance

1. Corn Insects. Only 3 of 100 inbred lines developed by State and Federal corn breeders from the Southern Corn Improvement Conference area had a satisfactory degree of resistance to first-brood European corn borer at Ankeny, Iowa. Six of 35 Southern inbred lines indicated a satisfactory degree to second brood resistance.

Translocation corn stocks were used to determine which chromosome arm(s) of the resistant inbred line B49 carries gene(s) for resistance to corn borer leaf feeding. These results indicate that B49 possesses a gene for resistance on the short arm of chromosomes 1, 2, and 4; and on the long arms of chromosomes 4, 6, and 8.



A study was initiated in 1964 that involved 3 permutations of each of 12 double cross hybrids and their nonparental single crosses. The double crosses (containing 0 to 4 European corn borer resistant lines) were made up of some combination of 8 inbreds (B49, C.I.31A, HD225, and B46 = resistant; B14, WF9, B37, and M14 = susceptible). Previous investigations have indicated that B49 and C.I.31A contribute a higher degree of resistance when in crosses with susceptible lines than HD225 and B46, and that susceptible B14 exhibits some degree of tolerance when in hybrid combination. Double crosses with B49 and C.I.31A plus either one or both of B46 and HD225 were rated resistant. Double crosses with B49 and C.I.31A with 2 susceptible lines rated as intermediate or better. Double crosses with both B49 and B46 were no better than when B49 was the only resistant line in the pedigree, and these crosses rated intermediate for leaf feeding. The double cross with B46 as the sole source of resistance was as susceptible as the double cross made up of 4 susceptible inbreds. The permutations of some of the double crosses differed from each other. This would be an indication of some degree of non-additive gene action being an operative force in resistance to corn borer leaf feeding.

In a greenhouse experiment substances were added to a resistant line, C.I.31A to see if it could be made susceptible. Four different treatments (1. vitamins plus dextrose, 2. vitamins, 3. dextrose, 4. distilled water) were injected into the plant through the side above the growing point. This treatment often caused an adverse effect on the plants, even when distilled water was injected. The vitamin plus dextrose treatment was not significantly different than the distilled water treatment. Dextrose alone was as good as vitamin plus dextrose, but vitamins alone had some detrimental effect on larval survival.

A few WF9 selections that originated from the breeding program of transferring corn borer resistance into WF9 germ plasm were evaluated when crossed onto 4 inbred testers. The original WF9 was the most susceptible entry in the test. Although the WF9 selections do not possess a high degree of corn borer resistance when in hybrid combination, they do appear to contribute some resistance.

In another test a commercially available fertilizer (which is readily soluble in water), amino acids, vitamins and dextrose were injected into corn hybrids or inbreds to determine their effect on larval survival and development. On the resistant corn, C.I.31A X B49, the survival was low on all treatments, but the check treatments were the lowest followed by the soluble fertilizer and amino acid treatments. Some of these treatments gave significant increases in larval survival, but the differences were not of the magnitude necessary to account for resistance. There were no significant differences between treatments on the susceptible cross, WF9 X B37, except the treatment of soluble fertilizer plus dextrose, which had a significantly lower survival than the other treatments.

The results from the resistant C.I.31A material were not very conclusive because of the low survival under all treatments. Vitamins, amino acids plus dextrose, and vitamins plus dextrose gave the highest survival on susceptible

WF9. The distilled water treatment was not significantly different from the untreated check.

Another study in 1964 involved plant juice (by use of a plant press) from the whorl area of C.I.31A and WF9. The "juice" of C.I.31A was put into the whorl area of WF9 plants with a hypodermic needle, and the juice of WF9 plants was put into plants of C.I.31A. There were no significant differences between treatments. However, the larval survival in this test was very low.

In a greenhouse test in late 1964 all of the ingredients of the corn borer synthetic diet except agar, leaf whorl, and cholesterol were applied to C.I.31A with a long hypodermic needle into the whorl area at the rate of 5 cc of solution per plant treated 3 times at 3-day intervals. Plants were dissected 10 days after egg hatch. Larval survival was 20 times greater on the "diet" treatment than on the distilled water.

B64 a yellow dent inbred tolerant to western corn rootworm developed cooperatively by USDA's Agricultural Research Service and Iowa Experiment Station was released to plant breeders in 1965.

Several hundred lines of corn were evaluated for corn rootworm resistance in South Dakota.

Laboratory techniques have been developed to test corn in the seedling stage for antibiosis to the larvae of the western corn rootworm. Evaluation is based on percent of original infestation recovered and size of recovered larvae after a given period. Differences have been observed based on the above criteria. Progeny tests are being conducted to evaluate these as potential sources of antibiosis.

Greenhouse tests with 36 inbred lines of corn revealed that three, R168, B55, and My3, possess a rather high degree of corn leaf aphid resistance.

Field cage studies at Tifton, Ga., showed that fewer earworm larvae could establish on unpollinated silks than on pollinated silks, indicating that either pollen itself, or the physiological or physical changes in silks initiated by fertilization helped larvae to become established.

Although slitting the husk of a resistant sweet inbred increased earworm damage, some factor or factors other than husk tightness contribute to resistance. Injury was not as great in resistant lines with slit husks as in susceptible inbreds with slit husks.

Six Southern Grain Insects Research Laboratory inbreds have indicated good resistance and quality in a hybrid testing program. Backcrosses of the inbreds in Walter's White have shown increased inbred vigor and maturation, yet little of the earworm resistance or quality has been sacrificed.

A feeding arrestant-stimulant obtained from lyophilized plant material was tested on filter paper for preference by larvae of the earworm, the fall armyworm, and the budworm. The response varied among the six species of plants tested, the plant portion used, and the species of insect. All three insect species preferred extracts of plant fruiting bodies to extracts of vegetative parts, indicating more arrestant in fruiting bodies. The feeding response was directly correlated to larval utilization and development, determined by incorporating plant material into diets. Utilization ranged from 12% to 61%.

A corn earworm larval feeding stimulant-arrestant extracted from corn kernels did not break down appreciably when stored dry at  $-10^{\circ}\text{C}$  for 9 months. It does lose much of its effect when reconstituted and held at room temperature for 48 hours. The stimulant was still active after the extract was heated to  $90^{\circ}\text{C}$  for 5 minutes. Heating separated out the inactive water-suspended portion. In dilute form the extract is more effective as an arrestant than as a stimulant.

An olfactometer with an electronic system consisting of a scanner relay with a miniature proximity sensor was used to monitor adult corn earworm flight and oviposition responses to chemicals, especially extracts from corn plants. A chemical fraction in corn kernels has been found to act as an oviposition repellent but chemical extraction and stability has been difficult.

In Mississippi silks of corn hybrids rated as resistant, intermediate, and susceptible to the corn earworm were analyzed. Twenty amino acids among the protein samples were identified with little or no differences among the three types of corn. The non-protein samples showed that there were 22 in each class of silks and that the concentrations were lower in the resistant lines, slightly higher concentrations in the intermediate, with the highest concentrations in the susceptible lines of corn.

Determinations of reducing sugars in susceptible dent corn showed 22.53% present in fresh silks and 15.03% present in the resistant single cross.

In Mississippi 239 hybrids and varieties were rated for corn earworm damage. Embro Departure VIII, PAG Experimental 15307, Miss. 6133, Funk's G707, Dixie 18, McCurdy M306, Dixie 55, Coker 911, Coker 811A, Coker 67, Coker 71, and Embro 256 CP were the most resistant entries.

A husk extension of more than 2 inches and a silk channel diameter of less than 1 inch gave maximum mechanical protection against earworm injury.

Seven single cross tests involving 54 inbred lines showed that 11 inbreds, Mp462, Mp468, Mp464, Mp335, Mp1, Mp420, Mp426, F6, Mp313E, L501, and Ab18 had a high degree of earworm resistance.

In tests at Lafayette, Ind., there were 27 experimental hybrids more resistant than the most resistant commercial hybrids.



2. Small Grain and Sorghum Insects. At Tifton, Ga., eight varieties of barley commonly grown in the Southeastern United States were compared with each other for their relative ability to resist attack by four species of aphids. Length of life span of the female and the number of progeny were used as criteria of evaluation. The decreasing order of resistance of each variety to each species of aphid is as follows: The English grain aphid, Will, Gajet, Early Marconee, Davie, Davie X Harbine, Dayton, Colonial 2, and Rogers; the apple grain aphid, Will, Early Marconee, Rogers, Gajet, Colonial 2, Dayton, Davie, and Davie X Harbine; the greenbug, Robers, Davie, Dayton, Davie X Harbine, Gajet, Early Marconee, Will, and Colonial 2; the corn leaf aphid, Dayton, Early Marconee, Davie X Harbine, Gajet, Colonial 2, Davie, Rogers, and Will.

At Lafayette, Ind., barley selections were screened for corn leaf aphid resistance under greenhouse conditions. Two selections, CI 1267 and CI 1417, from the barley variety Squarehead and one selection, CI 1365, from the variety Bolivia appeared to be immune to corn leaf aphids. The nature of the resistance of these three barley selections seemed to be non-preference and possibly some antibiosis, since the aphids did not feed or reproduce on the selections.

Populations studies of the 4 races of Hessian fly have been continued in Indiana. Race B which is capable of infesting the W38 resistant wheats Monon, Redcoat, and Reed increased considerably in 1964-65. Races C and D which are capable of infesting Knox 62 have increased slightly. This increase in Hessian fly populations capable of infesting the presently available wheat varieties make it desirable to release wheats having the Ribiero source of resistance as soon as possible. Hessian fly populations continue to decline in Kansas and Nebraska probably due to the increased use of resistant wheats especially Ottawa.

Temperature above 70° F affects the expression of resistance in W38, PI94587, and Ribiero derivative wheats. The average percent of infestations for W38 derivative wheats at 60, 70, 80, and 90° F were 1.3, 4.0, 5.3, and 14.7 respectively. While PI94587 reacted resistant at all temperatures, its derivatives Knox 62 and Dular had an infestation of 14.0% at 90° F. Ribiero and its derivative 4217 were influenced by high temperatures, while the derivative 5273 had no infestation at any temperature level. Ribiero and 4217 had average infestations of 4.0, 9.0, 57.5, and 100% at 60, 70, 80, and 90° F respectively. Increasing temperatures also had an effect on larval survival in susceptible wheat plants. At 60, 70, 80, 90° F the average number of flax seeds found per plant were 49, 35, 24, and 2 respectively. High temperatures may be useful in screening for different sources and level of resistance. By infesting wheats at high temperatures those wheats having high sources of resistance may express themselves better than those having a low level of resistance.

At Lafayette, Ind., 24 F<sub>2</sub> families from crosses between 21 Chinese monosomics and the fly resistant variety Ribiero were evaluated for resistance to Race A.

Four chromosome families segregated in ratios that would be expected if the critical chromosome were involved, these being 2D, 4A, 6B, and 7A. Cytological investigation of the susceptible plants showed none to be nullisomic. This is contrary to what would be expected if the critical chromosome were involved and indicates the above chromosomes may not be involved.

Monon wheat heads from approximately 500 flag leaf infested with cereal leaf beetle and 500 uninfested were analyzed for kernel weight, kernel number, protein content, pearling index, mill yield, and water retaining capacity. Results indicated an estimated 25% yield reduction but there were no significant losses in wheat quality.

At Lafayette, Ind., Hessian fly resistant selections were obtained from back-cross lines involving Genesee and the Hessian fly resistant wheats Redcoat, 4217, and 4835. Approximately 3,000 head selections, hybrids or lines from the Purdue regular fly nursery, preliminary yield nursery, advanced yield nursery, fly stem nursery, and from previous fly tests were infested with greenhouse populations of 1 to 3 races of Hessian fly. All wheats have one or more of the W38, PI94587, Ribiero, and Marquillo resistances in their parentage. Many Hessian fly resistant winter type Ribiero and PI94587 selections were made. With a build up of Race B in certain areas in southern Indiana emphasis is being placed on breeding wheats having the Ribiero and PI94587 types of resistance or combinations of these with the W38 or Marquillo resistance.

The Triticum species collection (581 entries), the Sando collection (842 entries) and the spring wheat collection (1,870 entries) grown at Galien, Mich., for cereal leaf beetle evaluation were rated for Hessian fly resistance also. Under a heavy infestation of Hessian fly in the area, 193 entries reacted resistant to Hessian fly.

Two hundred and five Tennessee wheats were evaluated by Races A and B to determine the type of resistance involved in some of their breeding material with unknown pedigrees. All the lines that reacted resistant to Race A reacted susceptible to Race B indicating the W38 resistance to be present in their breeding material. Five hundred and forty-seven entries of new wheat introductions (PI268825 - PI288033) were tested to Race A. One hundred and eighteen that reacted resistant were retested to Race D to determine if the Ribiero or complete PI94587 resistances were involved. Eighty-six of the lines resistant to Race A also reacted resistant to Race D.

Several promising wheat stem sawfly spring wheats are in advance yield nurseries in North Dakota. The most resistant were selections 60-54, 51-3549 x II-50-17, 61-107, and (II-50-17 x 51-2688) ND4-Rescue. Studies in North Dakota showed that wheat stem sawfly sex ratio differ for resistant and non-resistant lines of spring wheat, as host plant resistance increased, the percent of male progeny also increased. Indirect evidence was obtained which indicates that the female adult sawfly can selectively oviposit fertilized or unfertilized eggs and that host plant stimulation encourages oviposition of fertilized or unfertilized eggs.



Over ten thousand small grain accessions were evaluated for cereal leaf beetle resistance in a field nursery in Galien, Mich. Seven hundred and forty-two of the wheat entries had none to a trace of larval feeding and were considered as resistant. None of the oat or barley entries were found to possess a high level of resistance, although some entries seemed to be less preferred. Laboratory tests utilizing adult feeding and egg-laying (both the total number of eggs laid and location of oviposition) as criteria for measuring host-plant resistance have shown that: (1) the pubescence of certain wheat plants acts as a deterrent to oviposition, (2) some wheat plants cause either a cessation of egg-laying or a substantial reduction in the number of eggs laid, and (3) adults were capable of feeding upon all plants tested.

One hundred entries of the preliminary yield and advanced yield nursery having Hessian fly resistance were evaluated for cereal leaf beetle resistance. Three entries from the preliminary yield nursery and one entry from the advanced yield nursery had light to intermediate feeding by the cereal leaf beetle under a heavy beetle infestation.

#### H. Insect Vectors of Diseases

1. Corn Insects. Cooperative research by State and Federal entomologists and pathologist have shown that the disease infecting corn in the South and Midwest since 1962 is due to two separate viruses. The one in the South is a persistent leafhopper transmitted virus known as corn stunt while the one in the Northern Area is a non-persistent virus now named maize dwarf mosaic and can be transmitted by aphids and also mechanically. There is evidence that the two diseases may overlap in Missouri and Arkansas. Cooperative research by State and Federal workers at Wooster, Ohio, established that the corn leaf aphid can transmit maize dwarf mosaic. At Tifton, Ga., Dalbulus maidis, a known vector of corn stunt disease, was collected on volunteer corn from August 25, until the first killing frost on November 15, 1964.

Under greenhouse conditions at Tifton, 138 transmission tests were attempted using leafhoppers, other than D. maidis collected from corn growing in fields with plants displaying symptoms of corn stunt disease. Eighteen genera and 22 species of leafhoppers were represented. No symptoms of corn stunt disease were observed in any test.

In Mississippi 33 species of leafhoppers were collected on corn during the 1964 growing season. D. maidis was collected in large numbers during the period August 18 to November 19, in Louisiana, Texas, and Mississippi. Corn stunt virus transmission trials were conducted with 8 common species of indigenous leafhoppers. Discolorations which were not typical corn stunt virus symptoms developed on corn following feeding trials with Graminella nigrifrons but no positive symptoms of corn stunt was observed.

At Tifton, Ga., studies were conducted on the developmental times for D. maidis, a known vector of corn stunt disease. At temperatures of 55° and 65° F eggs were not laid. At 75° F 5 days were spent in the egg and 15 days in nymphal stadia. There was little difference in developmental times between 80°



and 90° F. The shortest developmental time observed was 12.5 days, with 2.5 days spent in the egg and 10 days in the nymphal stadia. The sex ratio of all reared progeny in all tests approached 1:1. The nymphs are rather quiescent and remain on the plants. Below 75° F the adults also tend to be quiescent, often found in the whorls, particularly of younger plants. The adults become very active at 80° and 90° F, flying readily from plant to plant. The temperature extremes that D. maidis can withstand are not known but tests indicate that it can survive over a range of at least 70° F. In a plant growth chamber one week survival was secured at a constant temperature of 102° F at both 50% and 80% relative humidity. Infested corn plants two weeks old were held at 32° F on three consecutive nights for 15 hours each and then returned to the growth chambers for observation. No mortality was noted and little, if any, effects were noted on subsequent development and reproduction potential.

Attempts to establish D. maidis on hosts other than corn in both growth chambers and in the greenhouse have been unsuccessful. These plants included several varieties of oats, wheat, barleys, rye, and sorghum. They can live and reproduce on Johnsongrass and gamagrass.

In connection with a research contract in Mississippi, a study was made of the predominant species of leafhoppers in the corn stunt area. Twenty-five different species were collected. G. nigrifrons was the most common species. Microlestes fascifrons was abundant in February and March, but was seldom collected in April and May, whereas the opposite was true of S. bicolor.

Research conducted in Israel under P. L. 480 project A10-ENT-5 on the rough dwarf virus disease of maize has shown that the virus can be maintained in the insect vector by transovorial passage to succeeding generations, even though the insects do not have access to host plants susceptible to the virus.

2. Small Grain Insects. At Brookings, S. D., barley yellow dwarf virus was transmitted by English grain and corn leaf aphids fed on extracts from infected barley using techniques previously developed for artificial feeding of the corn leaf aphid. Preliminary results have shown rate of centrifuging of crude plant extracts, acquisition feeding period, vigor of aphids, and population density of aphids in feeding cages are important influences on percent infectivity. The technique provides a useful tool for future basic investigations.

Studies on the effect of barley yellow dwarf on wheat yields indicated a reduction in winter wheat and 45% reduction in spring wheat.

Additional information on environmental conditions and techniques have permitted the testing of more than one hundred lines in the greenhouse for resistance to barley yellow dwarf virus. A few lines, mostly from New Zealand, have shown promise in maintaining productivity when infected with barley yellow dwarf.

The screening of more than 1800 lines of wheat for resistance or tolerance to barley yellow dwarf under field conditions, where healthy plots of each entry were grown adjacent to the diseased plots indicate that some lines possess resistance or tolerance to the virus or that they are symptomless carriers of the virus. Preliminary scorings of the entries show 1481 susceptible, 107 with mild symptoms, 209 with severe symptoms, and 14 with no symptoms.

At Manhattan, Kans., a migratory form of the wheat curl mite, the vector of wheat streak mosaic, has been observed in mite populations under laboratory and field conditions. These migratory forms are hardier, disperse more rapidly over plants, and distribute the eggs more widely than do the non-migratory mite forms. The dispersal of these migratory forms over wide areas when suitable hosts are present probably results in the development of infestations in epidemic proportions. Of all of the small grains, wheat continues to be the preferred host of the wheat curl mite.

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RICE INSECTS  
Entomology Research Division, ARS

Problem. Several species of insects including leafhoppers, the rice stink bug, rice water weevil, grape colaspis, stalk borers, and the sugarcane beetle damage rice in the rice growing areas of the United States. Progress has been made toward the solution of some of the insect problems encountered in the production of rice but more effective, more economical, and safer insect control measures are needed. The appearance of resistance to certain insecticides in some rice insects stresses the need for basic information to overcome this problem. Additional emphasis should be given to new approaches to control rice insects and to evaluate rice varieties for resistance to major rice insects.

USDA AND COOPERATIVE PROGRAM

The Department's program on rice insects involves entomologists, agronomists, and plant breeders, and plant pathologists engaged in both basic studies and in the application of known principles to the solution of growers' problems. The research is being conducted at Baton Rouge, La., in cooperation with the Louisiana Agricultural Experiment Station.

The Federal scientific effort devoted to research in this area totals 2.2 professional man-years. Of this number 0.6 is devoted to basic biology of the leafhoppers, rice stink bug, and rice water weevil; 0.5 to insecticidal control of rice stink bug and rice water weevil; 0.7 to varietal evaluation of rice for resistance to stink bug, rice water weevil, and vectors of rice diseases; 0.2 to insect vectors of hoja blanca and 0.2 to program leadership. In addition Federal support for research in this area provides for 0.4 man-year in a research grant to the University of Arkansas for varietal evaluation of rice for resistance to the rice water weevil.

PROGRAM FOR THE STATE EXPERIMENT STATIONS

Research on rice insects by the States is concerned with biology, ecology, and control. Studies are in progress which are designed to determine the economic importance of the various pest species present in growing rice. Efforts are being made to determine the amount of damage caused by different population levels of injurious species. Life history studies are under way which may reveal the vulnerable links in life cycles which can be exploited in control.

Information is being obtained on pest overwintering sites, spring emergence patterns, mating, egg deposition, length of developmental periods, food habits, number of generations per year, movement and dissemination and host relationships.

Biological and chemical control studies are being conducted in the field and laboratory to determine the most efficient and economical method for reducing damage. Rice samples from plots treated with insecticides are subjected to residue analyses.

There are 3.0 professional man-years dedicated by the States to research on insects affecting rice production.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAM

##### A. Basic Biology, Ecology, and Nutrition

Sperm were not transferred when male rice water weevils from Louisiana were mated with females from California. Weevils remained in a copulating position for 12 hours. Females that had been in copula for more than five minutes were dissected immediately after breaking copula and examined for the presence of sperm in the bursa copulatrix or spermatheca. A second group was dissected 24 hours after copulation. No sperm were found in either the bursa copulatrix or the spermatheca. Only female weevils have been found in California.

Rice water weevils confined on small rice plants in the laboratory at Baton Rouge, La., laid eggs in the leaf sheaths but none in the rice roots. Plants with eggs laid in the leaf sheath were also collected from the field at Crowley, La. The eggs were inserted paralleled to the plant fibers in the Perenchyma of the sheath, and the openings were plugged with a gelatinous substance. Egg deposition in sheaths usually occurred just below the water line.

The winter mortality of rice water weevils hibernating in grass clumps in Louisiana in 1964-65 was approximately 50%. None of the hibernating females examined in any of the samples contained sperm in the spermatheca.

An infestation of the lesser cornstalk borer, Elasmopalpus lignosellus was found in a rice field in Louisiana; this occurrence was the first record of this insect on rice in Louisiana. The infestation was controlled when the rice was flooded.

Sogata furcifera, a planthopper found on native grasses in Louisiana, lived an average of 16.5 days on healthy rice but 44.5 days on hoja blanca diseased rice, indicating that the hoja blanca virus is beneficial to this insect.

##### B. Insecticidal and Cultural Control

Seed treatments with UC-10854, UC-21149, Bayer 39007, Coumaphos, GS-13005, MCA-600, triphenyltin hydroxide, diazinon, SD-9129, aldrin, or an aldrin-heptachlor mixture did not provide adequate control of rice water weevil in a field experiment in Louisiana. Treatment with Bayer 39007 at rates of 1.0 and 0.5 pounds, SD 9129 at the rate of 0.5 pound and MCA-600 at the rate of 0.5 pound per 100 pounds of seed caused significant reduction in plant stand,

some of which was due to the interaction of the herbicide propanil used for weed control and these insecticides.

Aldrin applied at the rate of 0.25 pound per 100 pounds of seed failed to control water weevils at 5 of the 6 locations tested in Louisiana and Mississippi, indicating wide-spread occurrence of aldrin-resistant weevils in the southern rice-growing area.

Bayer 25141 applied as granules at the rate of 2 pounds toxicant per acre gave 80% control of rice water weevils. There was a significant increase in the number of rice water weevil larvae in plots treated with either phorate or Bidrin at the rate of 2 pounds toxicant per acre.

In a greenhouse experiment, carbaryl at 1 pound per acre gave a significant better reduction in egg hatch and nymphal survival of rice stink bugs over malathion applied at 5 pounds per acre.

Insecticidal seed treatment on rice, for control of rice water weevil, was evaluated for phytotoxicity in drill-seeded and water-seeded tests in the greenhouse. The insecticides were applied as an acetone solution of technical material. SD-9129 at 1.5 pounds per 100 pounds of seed, aldrin at 3 pounds, CL-52160 at 2 pounds, CL-47031 at 1.0 pound, B-44646 at 1.0 pound, and Di-Syston at 1 pound per 100 pounds of seed showed no phytotoxicity in drilled or water-seeded tests. Methyl parathion at 1 pound, parathion at 1 pound, GC-6506 at 0.5 pound, fenthion at 0.5 pound, Baygon at 0.5 pound, and triphenyltin hydroxide at 0.125 pound, were phytotoxic in water-seeded tests.

In a greenhouse test rice seed were treated at the equivalent of 0.75, 1.5, and 3.0 pounds toxicant per 100 pounds of seed with SD-9129, a phosphate insecticide with systemic capabilities. Germination was reduced at the 3.0 pound level. Nine days after planting, hoja blanca virus-infective adults of Sogata orizicola were caged on the plants. Six hours after caging, 69, 89, and 88% of the insects were dead in the respective concentrations, and 100% were dead at all treatment levels after 24 hours. Hoja blanca symptom development was reduced 63, 88, and 86% respectively indicating the insects had been killed prior to transmitting the virus to the rice plants. However, all treatment levels of the insecticide were ineffective 24 days after planting.

#### C. Varietal Evaluation for Insect Resistance

Seedlings of rice varieties vary in their attractiveness to Sogata orizicola. In greenhouse experiments, fewer insects were observed feeding on Nilo 1, Nilo 2, Chino, and Nato than on Bluebonnet-50, Gulf Rose, and Palo Gordo 503. Nilo 1 and Nilo 2 were most resistant to egg deposition and Gulf Rose was the most susceptible of 7 varieties tested.

#### D. Insect Vectors of Diseases

An experiment was run in cooperation with plant pathologists to determine if a



difference in transmission rate exists between acquisition periods of infective females starved for 48 hours and infective females held on Nato rice for 48 hours. The starved insects were allowed acquisition periods of 1, 2, and 3 hours with hourly transfers. The unstarved insects were allowed 1 hour acquisition. Very little transmission occurred at these acquisition periods and there were no differences between the periods or the two insect groups.

Eggs from viruliferous females were excised from leaf tissue, surface sterilized with 0.1% Hyamine solution, and placed into aseptic test tubes with aseptic rice seedlings growing in an agar medium containing a modified Hoagland and Knop solution. The tubes were placed in a temperature cabinet at 28°C. and under 14 hours illumination. After hatching, the nymphs were allowed to spend two days of the first instar on the seedlings, then were transferred to clean seedlings in aseptic tubes by vacuum through a sterile 27 gage needle. The nymphs adhered to the tip of the needle, and dropped off upon the termination of the vacuum. Disease symptoms were observed on two seedlings that had been used only during the first instar. This indicates that S. orizicola may receive the virus transovarially, and can feed and transmit the virus as first instar nymphs.

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FORAGE AND RANGE INSECTS  
Entomology Research Division, ARS

Problem. Numerous insect pests that attack forage and range plants in various parts of the United States lower seed production, reduce the quantity and quality of forage crops, and decrease the abundance of range plants for the grazing of livestock. Certain insects are involved in the transmission of forage-crop diseases. Among the more important insect pests are grasshoppers, lygus and other plant bugs, stink bugs, seed chalcids, the alfalfa weevil, root borers, spittlebugs, leafhoppers, and a variety of aphids including the spotted alfalfa aphid and the pea aphid. A variety of insecticides is used to control these insects but they are often costly and may create residue hazards in meat and milk as well as adversely affect wild-life. There is great need for more efficient insecticides that can be applied on forage crops and range vegetation without leaving residues harmful to man or animals or that might harm bees and other pollinating insects. Increased attention should be given to the development of non-chemical control methods. The search for insect parasites, predators, and pathogens and ways to employ them effectively should be emphasized in research. The development of crop varieties which resist attack by insects offers economical and safe insect-control procedures. Forage crops should be evaluated for resistance to major insect pests and resistant germ plasm should be made available for use by the plant breeders in crop-improvement programs. Basic studies are also needed on the feeding habits of grasshoppers under different environments that affect the abundance of these insect pests. New approaches to control of forage and range insects, such as sterilization techniques and sex attractants, should be investigated.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program of basic and applied research on forage and range insects. Studies on varieties of alfalfa resistant to insects are cooperative with State and Federal agronomists and plant breeders, those on plant disease transmission by insects with plant pathologists, and research on insecticide residues with chemists. Grasshopper research at Bozeman, Mont., Mesa, Ariz., and Columbia, Mo., is cooperative with the respective State Experiment Stations. White-fringed beetle research is conducted at Gulfport, Miss. Biological control studies on armyworms and cutworms at Baton Rouge are cooperative with the Louisiana Experiment Station. Investigations on alfalfa insects are being conducted at Mesa, and Tucson, Ariz., Lincoln, Nebr., and Beltsville, Md., in cooperation with the Experiment Stations in these States. Research on clover and turf grass insects at Forest Grove, Oreg., is conducted in cooperation with the Oregon Experiment Station. Work on grass insects, plant disease transmission by insects, and insecticide residues at Tifton, Ga., is cooperative with the Georgia Experiment Station. Research on insecticide residues is conducted at Beltsville, Md., and at Yakima, Wash., in cooperation with the Washington Experiment Station. Studies on varietal resistance, insect vectors of plant diseases and grass insects at

University Park, Pa., is cooperative with Experiment Stations in 12 North-eastern States. Certain phases of the research on forage and range insects are contributing to regional projects W-37 (Natural Factors Responsible for Grasshopper Population Changes), NC-52 (Factors Influencing the Distribution and Abundance of Grasshoppers), W-74 (Seed Chalcids Attacking Small-Seeded Leguminous Crops), and S-55 (Alfalfa Insects).

Two contracts and 5 grants with State Universities and Agricultural Experiment Stations will provide additional research on insect biology, physiology, and nutrition, biological control, attractants and varietal resistance.

The Federal scientific effort devoted to research in this area totals 26.5 professional man-years. Of this number 5.0 man-years are devoted to basic biology, physiology, and nutrition, 5.0 to insecticidal and cultural control, 4.0 to insecticide residue determinations, 4.6 to biological control, 0.6 to insect sterility, attractants, and other new approaches to control, 0.9 to evaluation of equipment for insect detection and control, 5.1 to varietal evaluation for insect resistance, 0.2 to insect vectors of diseases, and 1.1 to program leadership.

In addition Federal support of research in this area under contracts and grants provides a total of 1.9 professional man-years. Of this total 0.3 is devoted to basic biology, physiology and nutrition, 0.2 to biological control, 0.5 to insect attractants and other new methods of control and 0.6 to varietal evaluation of insect resistance.

A P. L. 480 project, (E21-ENT-9), "Insect Vectors of Virus Diseases of Various Forage Legumes" is underway with the Research Institute of Plant Protection, Poznan, Poland.

#### PROGRAM OF THE STATE EXPERIMENT STATIONS

Studies on forage and range insects comprise an important part of the research program in the States. Populations of insects are being studied in relation to meadow composition, density, age, and crop sequence or type of range. Data on physical and biotic factors are recorded and analyzed to determine their relationships to insect abundance. Studies are being performed to determine the effects of environmental conditions on life cycles and pest insect physiology. Relationships between such factors as diapause and flight patterns to body fat content are under investigation. The mechanisms by which insects orient to their hosts are being determined by comparisons of the relative attractiveness of various odors, baits, flower and foliage colors, plant shapes and humidity gradients.

Chemical control research is being performed including the comparative effectiveness of materials, different application schedules, residue analyses and relationships to cultural controls such as crop rotation, fertilization and destruction of pest breeding sites.



Biological control research includes studies of the biology and ecology of predators, parasites, microorganisms with a view to increasing their effectiveness in reducing the abundance of pest insects or weeds. New biological agents are being introduced as quickly as their value is demonstrated.

In forage crops, studies are under way to develop plant varieties resistant to insect attack. Crosses of resistant and susceptible plants are being made to determine mechanisms of inheritance. Resistance found is being incorporated into agronomically desirable varieties. Research is also being performed on insect transmission of plant diseases.

The States' programs include 63.2 professional man-years of research on forage and range crops.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAM

##### A. Basic Biology, Physiology and Nutrition

1. Grasshoppers. Peak populations of the desert grasshopper (Trimerotropis pallidipennis pallidipennis) were higher in 1964 than in any year since 1959. Light damage to crops and desert vegetation was observed at several locations, and spring flights of the desert grasshopper were more frequent and in greater volume than in other recent years. The increase in population was apparently influenced mainly by favorable rainfall and desert vegetation conditions. However, the December population in 1964 was much lower than in 1963. The spring population in 1965 was drastically lower than in 1964, and much lower than expected in view of apparently favorable rainfall and vegetation conditions from mid-summer 1964 through the spring of 1965.

Grasshopper populations in rangeland habitats in Peebles Valley, Ariz., increased in 1964 following 3 successive years of population decline. Vegetation was more succulent and the vegetative cover slightly greater in 1964. Populations ranged from 1.15 per square yard in grass to 1.53 in Juncus meadow.

After 3 years of fairly stable and very low populations of grasshoppers, the grasshopper population in Arizona rose 194% in Conservation Reserve Land and 167% in 4 other habitats. Populations were highest in grassy and weedy field margins. Dissosteira carolina, Melanoplus sanguinipes, and M. packardii were the leading species.

In the spring of 1964 at Mesa, Ariz., adult females of M. sanguinipes developed from some of the eggs laid by unmated females in the fall of 1963. The eggs went through the usual winter diapause and hatched in May. The adults had deformed front wings and hind legs and none of them laid eggs.

At Mesa, Ariz., male-female ratios of 1:3, 1:1, 3:1 of M. sanguinipes adults were tested. As the ratio of males to females was increased, there was an increase in the longevity of females, the number of egg pods deposited, the

percentage of pods deposited normally below the soil surface, and the average number of pods deposited per female per day. The sex ratio apparently had no effect on the percent hatching or the number of eggs per pod.

Paired adults of M. sanguinipes maintained in 6 x 6 x 12-inch cages showed higher egg production, higher percent hatching, and greater longevity, at a density of 2 pairs per cage than at a density of 24 pairs per cage. When nymphs of this species were reared in cylindrical plastic cages 2-3/4 x 13 inches long, with screen ends, there was little difference in the percent of survival to the adult stage at densities of 2 nymphs per cage and 12 nymphs per cage.

The body measurements of M. sanguinipes were greater for first-generation adults that were collected from dry rangeland in Arizona and reared in the insectary on a succulent diet than on adults of the same generation that completed their nymphal growth on the dry rangeland. Second-generation adults that were reared in the insectary on the favorable diet had larger body measurements than first-generation adults that received the favorable diet only during the latter part of the nymphal period.

On range recovery plots in Arizona the average grasshopper population on a sparse grass area for the period April-July 1964 in untreated fenced and grazed plots were 6.8 and 2.3 per square yard, respectively. By July 16, the percentages of grass eaten by one grasshopper per square yard in the fenced plot, with a grass cover of 7.7%, were 5.5 on blue grama, 4.6 on curly mesquite, and 14.1 on squirreltail. In October the percentage of grass cover was 15.5%, and in the fenced plot, it was 7.9% higher than in 1963.

At Bozeman, Mont., several species of range grasshoppers have been colonized in the laboratory, so they will be available for parasite and disease research. Schistocerca americana, brought in from Tennessee, is being reared successfully. Colonies of S. vaga vaga are being maintained for research on the adult diapause found in this species.

2. Alfalfa Insects. At Beltsville, Md., a non-diapausing colony of the alfalfa weevil was maintained in the laboratory under 8-10 hour day-length conditions for the third year. During 1964 six generations were completed. The effect of day lengths of around 18 hours of light, that normally yield diapausing weevils, was overridden by inserting a one hour dark period in the center. Larvae collected in the field during the spring and summer produced diapausing adults, while those collected in late fall produced non-diapausing adults.

Ninety-five modifications of an artificial larval diet capable of producing normal alfalfa weevil adults but less satisfactory than fresh alfalfa were tested at Beltsville with little improvement in survival and time required for development.



Previous cross mating tests between eastern and western strains of the alfalfa weevil demonstrated that they differ genetically and are partially isolated reproductively. Further tests during 1964-65 showed that when allowed free choice, males and females of both strains mated indiscriminately, egg hatch was reduced about 25%, and there was a slight increase in percentage of female progeny. When eastern and western males were placed with eastern females they competed equally as mates, egg hatch was reduced about 70%, and the sex ratio of progeny was equal. Eastern females confined with western males again produced only infertile eggs.

At Tucson, Ariz., an effort to break the adult diapause in the Egyptian alfalfa weevil, Hypera brunneipennis, was successful. A lot of eggs was incubated at room temperature, and the larvae obtained were grown to maturity on potted plants in a chamber in which 8 hours of light were provided each day. Seventy-three  $F_1$  adults emerged between May 20 and June 9. These were caged on potted alfalfa plants in the chamber under an 8-hour day. First eggs from these adults were recovered on June 28, 39 days after the first  $F_1$  adults emerged.

In the fall of 1964 sexuales of the spotted alfalfa aphid, Therioaphis maculata, continued to spread into States adjacent to Nebraska. These forms are now found in Iowa and Wyoming. This, plus the fact that an apparently separate occurrence of a holocyclic strain was found in Wisconsin, indicates that overwintering as an egg in the northern United States may become the rule rather than an exception for this species.

At Mesa, Ariz., studies were made on the life history and behavior of Aceratagallia curvata and Acinopterus angulatus under greenhouse conditions. Both of these leafhoppers are important pests of alfalfa in Arizona and vectors of important virus diseases. The life cycle of both species is less than 40 days and they are capable of producing several generations a year exclusively on alfalfa.

3. Clover Insects. At Lincoln, Nebr., pea aphids, Acyrtosiphon pisum, taken from sweetclover in the fall of 1963 and reared in the greenhouse reproduced and survived decidedly better on sweetclover than on alfalfa. A culture of these aphids was established, after some difficulty, on alfalfa and the resulting generations survived equally well on alfalfa or sweetclover. During the fall of 1964 pea aphids were taken from alfalfa and sweetclover in the field and reared on their respective hosts in the greenhouse. Reproduction and survival were better when the aphids were transferred to the same species of host, upon which they had been reared. This may indicate the development of a biotype with host preference.

4. Grass Insects. Observations were made at Lincoln, Nebr., on a midge (Cecidomyiidae) which destroys the developing caryopses of smooth bromegrass. It appears that there may be two generations during the short time before the caryopses begin to mature. Pupation apparently occurs within the flume at the site of the destroyed caryopses. All stages of a parasite (Eulophidae) were



found in association with the midge with the percentage of parasitism quite high. Unfortunately, the damage has been done by the time the parasite destroys the midge.

At University Park, Pa., weekly sweep-net collections in a stand of timothy and one of brome grass during the spring and summer of 1964 showed virtually the same major insects in each. These included the plantbugs Trigonotylus ruficornis and Lygus lineolaris; the leafhoppers Endria inimica, Graminella nigrifrons, Macrosteles fascifrons, Latalus sayi, Aceratagallia sanguinolenta; the fulgorid, Delphacodes campestris, the meadow spittlebug, Philaenus spumarius, and the pea aphid Acyrtosiphon pisum. The major diptera comprised a complex of the genus Oscinella.

At Tifton, Ga., the spittlebug, Prosapia bicincta, that attacks and severely damages Coastal bermudagrass, has been successfully reared from egg to adult. Eggs were placed on millet sprouts, and newly emerged nymphs fed on the roots for 4-7 days after which they were transferred to Coastal bermudagrass in 4-inch pots in a greenhouse. From 50 to 80% of the nymphs survived transfer, established themselves on the roots, and made new spittle masses. The cycle from eggs to adults required 40-59 days.

A study was made to determine how the spittlebug damages Coastal bermudagrass. Adults were caged on single stems in 4-inch pots. An adult spittlebug was confined to single stems of Coastal bermudagrass for 1, 2, 4, 8, 16, 24, 48, 96, and 168 hours and the plants were observed for 2 weeks. Damage symptoms began to appear in 3 days and consisted of a yellow stippling of the leaves and sheaths. The stippled areas coalesced to form streaked areas at about 5-7 days. After 7-10 days the leaves turned brown and dried up. The number of leaves which showed symptoms and the severity of the damage was in direct proportion to the length of time the adults fed. When adults were confined to stems below the attachment of the first leaf sheaths, symptoms appeared in the leaves above and in the growing tip. When adults were confined on one branch of a forked stem, symptoms appeared only on the branch on which the insect fed. Observations indicated that the adults introduce a toxin into the vascular tissue that destroys the chlorophyll. Nymphal feeding did not cause similar symptoms. Stems fed upon by 4th instar nymphs had small dark brown lesions near the soil level. The lesions were strictly local because xylem and phloem appeared normal a few microns above the lesion. It is thought that the nymphs kill the cells in the immediate vicinity of the feeding site and that if a toxin is secreted it is not translocated.

Spittlebug eggs entered diapause when held at 80° F constant temperature and in water-saturated filter paper in plastic petri dishes. Photoperiods of 14 and 16 hours per day did not prevent diapause of newly laid eggs. However, allowing the filter paper and attached eggs to air dry one day, prevented diapause. Chemicals were found to break diapause. Best results were obtained with a 30-second dip in laundry bleach. Incubation of diapausing eggs at 100° F for 7 days and at 48° F for 7 and 14 days was 30-35% successful in breaking diapause.

At Tifton, Ga., spittlebug adults were taken in fluorescent light traps in large numbers. The traps ranged from 6 to 90 inches in height above ground. The proportion of females captured was less than 5% in the highest traps but increased as the height of the traps decreased. Sexes were about equal in traps 6 inches high located in a heavily infested area. Traps, even of the low type, nearest the periphery of an infestation caught less than 20% females. These observations indicate that adults migrate from heavily damaged areas and that the males move faster, or perhaps farther than the females. The gravidity of the female population as determined through dissection of 700 specimens, was not correlated with the proportion of females caught in relation to the height of the traps.

A study of overwintering bermudagrass sod reserves indicated that losses due to spittlebug damage in the fall is carried over into the spring with reduced yields of forage. The ability of Coastal bermudagrass to recover from severe damage also was demonstrated.

At Mesa, Ariz., two species of leafhoppers, Carneocephala nuda and C. triguttata, were successfully crossbred in captivity. Both species are important pests of bermudagrass. C. triguttata is a vector of Pierce's disease virus of grapes or alfalfa dwarf virus. All intraspecific, interspecific, and reciprocal crosses produced progeny in the F<sub>1</sub> generation.

At Forest Grove, Oreg., studies on several species of sod webworms indicate that Crambus topiarius is the principal economic species. In some years it has been very injurious in old plantings of Merion bluegrass. While it is prevalent in other varieties of bluegrass, as well as in the fescues and bentgrasses, it has done much less obvious damage to these grasses. The larvae feed both upon the grass blades and upon the root system, but the root feeding causes the most damage. C. bonifatellus, while usually scarce or entirely absent in commercial grass plantings, is common on lawns. Larval damage to the sod is usually not evident, possibly because bonifatellus prefers lawns that have been adequately fertilized and watered; hence the sod may be able to recoup damage losses as fast as they occur.

C. bonifatellus proved to be one of the most adaptable of the crambid species for rearing from egg to adult in the laboratory. C. dorsipunctellus is one of the most common species on open bunch grass rangelands and in fescue seed producing areas but its rank of economic importance is not yet established. C. leachellis and C. tutillus are two species that are occasionally abundant in certain very restricted areas. C. leachellis occurs in bent and mixed wild grasses in the rainy seacoast area as well as inland areas up to 2000 feet elevation in central Oregon. C. tutillus moths were found only in very limited areas and chiefly in the short fescues. No larval damage was noted that was caused by this species.

5. White-fringed Beetles. White-fringed beetle larvae Graphognathus peregrinus, severely damaged new plantings of pine seedlings in Mississippi, in January and February 1965. The seeds, planted in December 1964, were



treated with endrin as a bird repellent, which apparently did not affect the larvae. Seedlings were cut off just below the surface of the ground when they were approximately one inch in height. This indicates that these larvae can feed and cause economic damage during the winter.

During the spring of 1965, larvae of G. peregrinus severely damaged a field of sugarcane near Grand Bay, Ala. The principle damage was caused by the larvae feeding on the eyes or buds as they grew from the seed pieces thereby greatly reducing the stand. Over the entire field, stand reduction was estimated at 30% but was as high as 90% in certain parts of the field.

A grant was recently awarded to Auburn University to develop artificial rearing techniques for the white-fringed beetles, and research is being initiated.

## B. Insecticidal and Cultural Control

1. Grasshoppers. Small plot tests were conducted at Bozeman, Mont., to compare low-volume applications of several insecticides with an aldrin standard against rangeland grasshoppers. Undiluted MCA-600 at 4 and 6 ounces per acre (spray volume 2 and 3 pints per acre, respectively) and undiluted carbaryl concentrate at 4, 6, and 8 ounces per acre actual (spray volume 1, 1-1/2, and 3 pints per acre, respectively) approached but did not equal the standard in effectiveness. Naled diluted with light, cracked gas oil at 6 and 8 ounces per acre actual (spray volume 1-1/2 and 2 pints per acre, respectively) also approached but did not equal aldrin. Diazinon in post treating oil at 8 ounces per acre actual (spray volume 2 pints per acre) was more effective than the standard. American Cyanamid EI-47772 at 2 ounces per acre actual in post treating oil (spray volume 2 pints per acre) was well below the standard in this test.

Laboratory screening tests were conducted with 30 insecticides against adults of the migratory grasshopper, M. sanguinipes. Acetone solutions of the insecticides were applied topically at 2.10 and 10.50  $\mu$ g per insect. Only two compounds NIA 10242 and Dursban compared favorably with aldrin.

Several insecticides were applied by aircraft at ultra-low volumes. Bidrin at spray volumes of 7.4 and 8.4 ounces (0.93 and 2.10 ounces active material) per acre, gave satisfactory control. Application of 5.6 to 11 ounces of active material per acre gave mortalities ranging from 87.7 to 97.7%. Satisfactory control was obtained with diazinon at 8 to 10 ounces of active material per acre. Carbaryl in the formulation tested was too volatile; crystals formed in the spraying system and dosages were questionable. Bayer 25141 gave very good control at dosages of approximately 1 and 3 ounces per acre.

2. Alfalfa Insects. At Beltsville, Md., fall 1964 and early spring 1965 applications of commercial and experimental insecticides failed to give adequate control of the alfalfa weevil on first crop alfalfa. Thirteen experimental insecticides applied to the growing crop in the spring of 1965



gave weevil control equal to or better than methoxychlor at 24 ounces.

During the winter 38 new materials were screened in the laboratory against adult alfalfa weevils. Those showing most activity in residual tests were Bayer 39007, GC 6506, Hercules 14504, and Velsicol FCS-13. Materials showing a high degree of systemic action were: GC 3707, GC 6506, Stauffer R-7239, R-7240, and B-10288, Velsicol FCS-13, and OCS-21959, VC-3-668, and VC-9-104.

At Lincoln, Nebr., insect populations in alfalfa were generally low and adverse climatic factors were not favorable for good seed set. Plant bugs and alfalfa seed chalcids, both of which directly affect seed production, were present. Six insecticides applied in the prebloom stage were equally effective initially against the plant bugs but none equalled DDT in producing a lasting effect. None of the insecticides were effective against the alfalfa seed chalcid.

In three tests during 1964 and the first half of 1965 baits containing mirex in soybean oil impregnated in corncob grits showed little promise for control of the red harvester ant, Pogonomyrmex barbatus, in irrigated alfalfa fields near Mesa, Ariz. In some of the test series, the ant colonies had been treated for more than a year at the last inspection in the spring of 1965.

3. Grass Insects. At Tifton, Ga., several insecticides were tested during the summer of 1964 to control the spittlebug, Prosapia bicincta on Coastal bermudagrass. Granular lindane was superior to all other insecticides tested for control of nymphs and adults. Sprays of Bidrin and MCA-600, at 1.0 pound per acre were effective in reducing populations of nymphs while sprays of Imidan and Bomyl, at 1.0 pound per acre were not. Bomyl and diazinon granules, both at 1.0 pound per acre, showed promise.

At Tifton, Ga., insecticides were applied to Coastal bermudagrass to control several phytophagous insects including the fall armyworm, and a composite of leafhopper species. An increased yield of grass forage was obtained with sprays of MCA-600, Bomyl, and Bidrin at 0.5 pound per acre. Endosulfan, carbaryl, diazinon, and Imidan were effective in reducing the insect complex but no increased yield of forage was measured in these treatments.

At Tifton, Ga., the insecticide GC 3707 was applied to Coastal bermudagrass, silage corn, and millet in sprays at rates of 4 and 8 ounces per acre. Samples of foliage were taken to the laboratory and fed to 4-day-old fall armyworm larvae. Mortality counts were made at 24 and 48 hours. The 8-ounce rate gave immediate control of 98% on corn, 84% on bermudagrass, and none on millet. The 4-ounce rate gave 43% immediate control on corn and poor control on the other crops. Samples taken 1 day after treatment showed only negligible control with both rates on the 3 crops.

4. White-fringed Beetles. At Gulfport, Miss., overwintering larvae of G. peregrinus were placed in soil containing extremely high concentrations of dieldrin and Telodrin to determine if it would be feasible to control larvae

damaging new plantings of pine seedlings in January and February. No mortality occurred when these larvae were placed in soil containing 250 pounds of actual dieldrin per acre and 2000 pounds gave only 40% mortality. Telodrin at 250 pounds per acre gave only 60% control and 1000 pounds were necessary to give 100% control.

### C. Insecticide Residue Determinations

1. MCA-600 Residues on Silage Corn. At Tifton, Ga., MCA-600 was applied to corn at 8 ounces per acre in a spray. Analysis of samples of corn taken the day of treatment and at intervals of 1, 2, 4, 7, 14, 21, and 28 days showed residues on a green weight basis of 7.46, 6.40, 2.85, 1.21, 0.53, 0.16, <0.10, and <0.10 ppm, respectively.
2. Bidrin Residues on Silage Corn. At Tifton, Ga., Bidrin was applied to corn at the rate of 4 ounces per acre in a spray. Samples of corn were taken the day of treatment and at intervals of 1, 2, 4, 7, 14, and 21 days were analyzed at Beltsville, Md., and the residues on a green weight basis were, respectively, 6.32, 1.52, 0.64, 0.46, 0.18, and 0.10 ppm. The minimum sensitivity of the analytical methods was 0.1 ppm.
3. Imidan Residues. Standing corn was sprayed with Imidan at 4, 8, and 16 ounces per acre, harvested 24 hours later, chopped and packed in gallon jars. Samples of the chopped corn were analyzed for residues and after 59 days the jar silage was analyzed for Imidan residues. The residues found were as follows: 4 ounce per acre chopped corn 2.36 ppm and silage 1.06 ppm, 8 ounces per acre chopped corn 5.32 ppm and silage 1.77 ppm, 16 ounces per acre chopped corn 14.72 ppm and silage 4.97 ppm. The loss of Imidan in the 59-day storage period ranged from 55 to 67%.

Standing corn grown for ensiling was treated with Imidan spray at 4, 8, and 16 ounces per acre, and sampled immediately after treatment and at intervals of 1, 2, 4, and 7 days. The samples were analyzed by the total phosphorous method. Residues in ppm at the above stated intervals were as follows: 4 ounces per acre, 9.23, 2.36, 1.69, and 1.36, (the 7th day sample was lost); 8 ounces per acre, 15.57, 5.32, 4.79, 3.82, and 2.30; 16 ounces per acre, 17.16, 14.72, 13.67, 10.67, and 7.20.

At Tifton, Ga., Imidan was applied to silage corn at 8, 16, and 32 ounces per acre as an emulsion spray. The plants were sampled as soon as the spray was dry, and one day after treatments. The initial residues obtained with the 8, 16, and 32 ounce treatments were 6.80, 11.36, and 30.0 ppm, respectively, and one day after treatment, when the corn was ensiled, the residues were, respectively, 3.43, 9.72, and 20.4 ppm. During approximately one month's storage these residues degraded to 1.56, 6.27, and 10.02 ppm. The silage was fed dairy cows for 50-56 days after the month's storage and during the feeding period silage samples composited weekly averaged 1.10, 3.42, and 6.88 ppm, for the 8, 16, and 32 ounce treatment, respectively. Milk samples were analyzed by gas liquid chromatography using an electron affinity detector.



No residue was found in excess of the minimum sensitivity of the analytical method. Whole blood samples taken from the cows at weekly intervals showed no evidence of cholinesterase inhibition in animals fed corn silage treated at rates of .5 or 1 pound per acre. There was evidence of a minor (less than 20%) inhibition of cholinesterase in animals fed the 32 ounce treatment for a period of 56 days.

4. Dimethoate Residues in Silage. At Beltsville, Md., samples of alfalfa that had been sprayed with dimethoate were collected before ensiling and at intervals afterwards. Samples collected before ensiling contained from 81.6 to 95.8 ppm of dimethoate. The residues in silage samples decreased as the experiment progressed, falling from an average of 41.7 ppm to 25 ppm over a period of 2 to 2-1/2 months.

5. Phosphate Insecticide Residues on Alfalfa. Samples of alfalfa at Beltsville, Md., were analyzed from plots that had been treated with 1 pound of Imidan, 1 or 0.25 pound of Bayer 25141, or GC 3707 at an unstated dosage. The samples were taken 35 days after treatment. No residues of GC 3707 were detected in excess of the sensitivity of the analytical method (0.06 ppm). Residues of Bayer 25141 averaged 0.47 ppm from the 1-pound per acre plot and 0.16 ppm from the 0.25-pound plot. A residue of 0.16 ppm of Imidan was found in the alfalfa treated with that insecticide.

6. Heptachlor Residues. At Beltsville, Md., samples of soil and alfalfa were taken in the spring of 1964 before the first cutting of alfalfa from alfalfa fields that had been commercially treated with heptachlor in the fall of 1963. Analysis of the samples showed the presence of 0.16 to 0.25 ppm of heptachlor epoxide in the alfalfa from treated fields. The soil samples contained heptachlor residues ranging from 0.033 to 0.018 ppm and heptachlor epoxide residues from 0.004 to 0.085 ppm.

In translocation studies at Beltsville, Md., alfalfa plants were grown in porcelain pots in soil treated with heptachlor or heptachlor epoxide at rates of 1 or 4 pounds per acre. Three cuttings of the foliage were made and samples were analyzed for heptachlor and heptachlor epoxide. Results indicated that there was no translocation of heptachlor or heptachlor epoxide into the aerial parts of the alfalfa plant in amounts detectable by the method of analysis employed (minimum level 0.01 ppm).

7. Bayer 25141 Residues in Range Grass. Bayer 25141 was applied to range grass at the rate of 1, 2, or 3 ounces of active ingredient per acre. Samples of the range grass taken 0, 7, and 44 days after application and analyzed at Yakima, Wash., showed residues of 29.8, 19.4, and 4.6 ppm from the 1-ounce dosage, 54.0, 42.0, and 12.3 from the 2-ounce dosage, and 64.6, 53.9, and 19.2 ppm from the 3-ounce dosage.

8. MCA-600 Residues on Range Grass. Samples of range grass were taken before and at intervals after treatment with 2 or 4 ounces of undiluted MCA-600 per acre. Residues found at 0, 7, 14, and 28 days after application



were 37.8, 21.4, 12.0, and 2.4 ppm for the 2-ounce application and 58.8, 52.1, 35.6, and 7.9 ppm for the 4-ounce application.

9. Phorate Residues on Lupine. At Tifton, Ga., biological assay indicated that there were residues of phorate in seeds of yellow and blue lupine harvested from fields treated with a granular formulation. Granular phorate is used to control aphids thus reducing the incidence of Bean Yellow Mosaic Virus and increasing seed production.

#### D. Biological Control

1. Grasshoppers. In 1964 natural enemies, mostly birds and dipterous larval parasites, were abundant and destroyed many grasshoppers in limited areas in Arizona. In June, on the San Carlos Apache Indian Reservation, parasitism of adult grasshoppers by larva of flesh flies was 2.9% and ranged up to 9.0%. In mid-October, in a small area bordered by oak trees, adults of Trachyrhachys mexicana and Arphia p. crassa were parasitized 66.6 and 57.1%, respectively, by larvae of Trichopsidea clausa. By mid-September on rangeland in extreme southern Arizona adults of Morseilla flaviventris and Boopedon nubilum grasshoppers were parasitized 60 and 36%, respectively. There were very few mite-infested grasshoppers on rangelands in southeastern Arizona.

At Columbia, Mo., studies on the red grasshopper mite, Eutrombidium trigonum revealed a method of sperm transfer involving the production of spermatophores. The spermatophores consist of an orange-colored capsule attached to the apical portion of a thin, translucent, whitish stalk whose base is embedded in the substratum. The capsule enclosed a homogeneous mass of granular material. No active sperm were observed in the capsules examined. The spermatophore stalk from the base to the branch supporting the sperm capsule averaged 318 microns in length. The diameter of the capsule averaged about 75 microns. After deposition of the spermatophore the male mites invariably performed a nuptial dance before resuming their previous exploratory or feeding behavior. This is the first instance in which the production of spermatophores by male mites has been found in this genus, although other members of the family Trombidiidae have been known to produce spermatophores.

During the 1964 season field applications of spores of the protozoan, Nosema locustae were made in 9 locations in northwestern Montana. Except for one site, the spores were applied in a localized area, generally no larger than 100 square feet. Two months after application infection rates at these 9 sites ranged from 10 to 50% at the application points. The percent of infection was less at points away from the area where the spores were applied. No significant infections were found in untreated control sites. From this it was determined that it may be feasible to apply Nosema spores under field conditions.

The field study for observing the disease under natural conditions is being continued in Camas County, Idaho. During 1964, more than 4% of the grasshoppers examined were infected with detectable levels of Nosema spores. This

level approximates that observed during 1963; however, since grasshopper population levels were considerably higher during 1964 the actual number of infected specimens was double that of 1963. At four of the plot locations the average infection during September was in excess of 50% and as high as 100% at one location.

2. Alfalfa Weevil. In 1965, the alfalfa weevil continued to spread in Arkansas, Missouri, Illinois, Indiana, Ohio, and Vermont. Releases of parasites were continued in Vermont, New York, Massachusetts, Delaware, Pennsylvania, Ohio, Indiana, Illinois, Missouri, and Virginia. Releases included Bathyplectes curculionis, B. anurus, Tetrastichus insertus, and Microctonus aethiops. All of these are established and spreading at one or more locations. Parasitism by T. insertus at the 1962 release site in Massachusetts was 64% in the spring of 1965. Parasitism by B. curculionis at a Massachusetts release site was 44%. Parasitism at release sites in Pennsylvania, Maryland, and Virginia was less than 5%. Some build-up and spread of these two parasites are indicated.

A study was begun in the spring of 1963 to determine the amount and effect of parasitism on the alfalfa weevil in an area immediately east of the Rocky Mountains which includes the eastern parts of Colorado and Wyoming and the western portions of Kansas, Nebraska, and South Dakota. The weevil is not as damaging in this area, nor is it spreading nearly as fast as it is in the Eastern States. Parasitism by B. curculionis ranged from 8% to 65% in 1963. and from 13 to 96% in 1964.

3. Egyptian Alfalfa Weevil. Two parasites of the Egyptian alfalfa weevil, Hypera brunneipennis, were released in alfalfa fields near Mesa, Ariz., during March and April 1965. A total of 567 adults of Tetrastichus incertus, a larval parasite, was released March 9, and 743 larvae of Microctonus aethiops, an adult parasite were released March 18 and April 7. In connection with the parasite releases, seasonal abundance counts of H. brunneipennis larvae were made. Larvae were collected each week from February 2 to May 26. Peak populations averaging about 600 larvae per 100 sweeps were recorded March 23 and 30.

4. Spotted Alfalfa Aphid. Surveys in 1964 showed that Trioxys utilis was widely distributed in central and southern Arizona and southwestern New Mexico. No other parasite species were recovered. Of all aphids examined only 1.7% were parasitized and the highest in any field was 8%.

Observations throughout 1964 in four alfalfa fields near Mesa, Ariz., showed that the leading predators were Orius spp., Collops vittatus, and nabids. The total number of predators decreased 27% compared with 1963. The lady beetle population was very low until mid-March. Then it increased sharply to a peak in mid-April which coincided with the peak of the spotted alfalfa aphid. No aphid mortality from fungus diseases was observed in the four fields. Average parasitization by T. utilis increased slightly, from 8.54% in 1963 to 9.91% in 1964. Populations of spotted alfalfa aphids, lady beetles, and total predators were 65.4, 77.5, and 49.6% lower, respectively, than for the 6-year period 1958-63.



The decrease in lady beetle populations in Arizona for the 10 years, 1955-64, has coincided with a decrease in the spotted alfalfa aphid population. During the spring of 1965 the percentage of aphids parasitized by T. utilis averaged 21.9% compared to 10.0 in 1964.

5. Armyworms and Cutworms. At Baton Rouge, La., parasitism of the fall armyworm averaged about 40%, which is lower than previously recorded in these studies. Disease was non-existent in the collections.

6. Rhodesgrass Scale. In June 1965 a contract was awarded to Texas Agricultural Experiment Station, at Weslaco, Tex., to investigate means of mass producing and distributing Neodusmetia sangwani, an effective parasite of the Rhodesgrass scale.

#### E. Insect Sterility, Attractants, and Other New Approaches to Control

1. Spittlebugs. Females of the spittlebug, Prosapia bicincta, give off a strong vanilla-like odor during the first week of adult life. At Tifton, Ga., experiments were conducted to determine if the odor was a sex attractant. Virgin females were confined in one branch of a Y tube with Coastal bermudagrass as food. The other branch contained only Coastal bermudagrass. Field collected males were introduced at the base of the Y. A fan pulled air toward the base of the Y. Tests with females 1 through 8 days of age resulted in more males being collected in the food branch than the female branch of the Y.

2. Alfalfa Weevil. At Beltsville, Md., laboratory and field tests with liquid propane flamers demonstrated that applications of heat to established alfalfa fields in the spring before growth begins effectively controlled the alfalfa weevil without damage to the crop. Weevil eggs in stubble were more susceptible than adult weevils. Some weed control was also obtained. A new alfalfa seeding, however, was severely damaged by all levels of heat application.

Research is being initiated at Blacksburg, Va., under a grant awarded in June 1965 to Virginia Polytechnic Institute to investigate attractants and stimulants for the alfalfa weevil.

3. Alfalfa Seed Chalcid. In June 1965 a grant was awarded to the University of Wyoming to study the ovipositional behavior of the alfalfa seed chalcid to chemicals occurring in alfalfa.

#### F. Evaluation of Equipment for Insect Detection and Control.

1. Grasshoppers. At Bozeman, Mont., a miniature spinning cage atomizer, developed by the Plant Pest Control Division, and popularly known as the Mini-spin, was compared with standard boom and nozzle spray gear in tests for control of grasshoppers on rangeland. Forty-acre plots were treated with aircraft using undiluted technical malathion. The best control was obtained when treatment was made with the Mini-spin nozzle, but the differences did not prove statistically significant. Spray patterns taken on dye-cards showed the



best control of droplet size to be from the Mini-spin. The ultra-low volumes applied per acre were believed largely responsible for greater variation in the mortalities than usually found in similar field tests with larger volumes of spray material.

2. Alfalfa Weevil. An experimental field applicator was designed at Beltsville to deliver technical liquid insecticides in aerosol form in amounts as low as 4 ounces per acre. This was mounted on a tractor and the aerosol disbursed under a trailing plastic canopy. In field tests the aerosol application did not equal conventional spray applications for alfalfa weevil control. The canopy allowed some of the aerosol to escape.

#### G. Varietal Evaluation for Insect Resistance.

1. Potato leafhopper. A field evaluation was made at Lincoln, Nebr., on 75 alfalfa clones for resistance to the potato leafhopper, Empoasca fabae. Visual ratings were based on an estimate of the percent of leaf tissue which showed feeding injury. In general, higher leafhopper populations occurred on the more heavily damaged plants; however, equal numbers of leafhoppers did not cause the same degree of injury to all clones. Certain resistant clones appeared to be unattractive to leafhoppers; others appeared to be tolerant. Correlations between visual ratings and population density, as measured by nymphs per gram of plant material, were all significant at the 1% level.

At University Park, Pa., a detailed comparison of selected leafhopper-injured and sprayed alfalfa clones planted in pairs in a space nursery showed that in addition to yellowing, damaged plants were shorter, had a higher percent of dry matter, shorter middle and top internodes, and more branching than protected plants. The kinds of injury are being explored for possible use in measuring antibiosis reactions of alfalfa strains to potato leafhoppers.

The potato leafhopper is not considered to be a pest of sweetclover, but in 1964, certain experimental lines in a nursery at Lincoln, Nebr., showed considerable injury by this insect. Of the recommended varieties present, Denta showed no visual injury and injury to the others was light. There was a positive correlation between leafhopper populations and injury ratings. In a subsequent greenhouse test leafhopper reproductive rates were significantly higher on one of the entries that showed damage in the field than on two others which showed little or no damage. While all the recommended varieties had some degree of resistance, it should be noted that there are lines which do not have resistance. This may be important because the use of other characters from these lines could result in the transfer of leafhopper susceptibility to improved varieties.

2. Alfalfa Weevil. Improvements were made at Beltsville, Md., in laboratory testing techniques for the alfalfa weevil. Placing cut stems in flats of sand rather than in individual water vials reduced materially the time required for setting up test material for oviposition and feeding response. The preparation of material for larval survival tests was also reduced by placing cut stems in

agar slants in glass tubes. The use of seedling plants in adult feeding preference tests was found to be a promising method of rapid selection. Significant differences were found among alfalfa entries in terms of egg laying, adult visitations to stems, adult feeding on stems and seedlings, and larval survival.

3. Egyptian Alfalfa Weevil. From 18 alfalfas planted in rows at Yuma, Ariz., in 1963, 71 plants were selected for possible resistance to larval feeding of H. brunneipennis. Selection was made on the basis of superior growth and light feeding marks during a period of severe damage to the planting as a whole. Eighteen of the plants were selected after new-generation adults had fed heavily for several weeks, their superior condition indicating possible resistance to adult feeding as well.

At Mesa, Ariz., about 1000 plants were visually rated in field plots for resistance to this insect. After two years of field evaluation only 16 plants were selected which appeared to be free of damage.

4. Spotted Alfalfa Aphid. At Tucson, Ariz., 442 Sonora and Moapa clones selected for possible resistance to alfalfa mosaic on the basis of tall stems and healthy roots were given stem-cage tests to determine level of resistance to spotted alfalfa aphid. The testing was performed in cool weather of winter and early spring to permit isolation of the clones with resistance that would hold up the year round. Forty-five clones were classified as highly resistant.

Twenty-eight clones selected at Mesa, Ariz., for resistance to the alfalfa seed chalcid were tested for resistance to two biotypes of the spotted alfalfa aphid. Sixteen were classified as highly resistant to both biotypes and two as susceptible to both. Most of the others were highly resistant to biotype ENT B but had lower or no resistance to ENT A.

Thirty-eight clones from the Nevada breeding program selected for good performance in a root disease plot at Mesa were tested for resistance to the spotted alfalfa aphid. Twenty-three were highly resistant to both biotypes.

Eighteen experimental and commercial alfalfa varieties were screened in the greenhouse for plants resistant to the spotted alfalfa aphid in the seedling stage. Four hundred and twenty-seven clones were selected from 48 flats of material. These were given stem-cage tests after being transplanted to the field and 292 were found to be highly resistant to both biotypes.

In greenhouse studies at Lincoln, Nebr., plants resistant to the spotted alfalfa aphid, Therioaphis maculata, that received a low level of Ca or a high level of N had a significantly higher number of live aphids after 4 days in comparison to resistant plants that received medium or high levels of Ca or low and medium levels of N. Nymphs produced and maintained on the terminal ends of low Ca resistant plants and high N resistant plants grew to maturity and reproduced. Susceptibility was not altered by any of the N or Ca levels. None of the other treatments (K, P, S, and Mg.) altered survival or reproduction of the aphids on resistant or susceptible plants.



At Manhattan, Kans., progress is being made under contract with the Kansas Agricultural Experiment Station in developing alfalfa varieties with combined resistance to two or more insects. Out of 187 plants from the synthetic variety KS10, 23 were resistant to the spotted alfalfa aphid, pea aphid, and bacterial wilt.

In connection with the above contract, studies were made on nymphal and adult survival of the spotted alfalfa aphid on excised and intact trifoliolates. Excised plant parts are frequently used in screening for resistance, particularly antibiosis. Of 50 clones tested, 24% showed significant differences in adult survival and 20% in nymphal survival comparisons. In all cases where the differences were significant, the survival on excised trifoliolates exceeded that of the intact trifoliolates of the same clone. These results indicate that using excised trifoliolates with the spotted alfalfa aphid might tend to underestimate the resistance level and make a clone appear somewhat more susceptible.

5. Pea Aphid. Seventy-eight Arizona clones resistant to root rots and the spotted alfalfa aphid were tested for resistance to the pea aphid. Eight clones derived from African and 10 derived from Lahontan were found to have a useful level of antibiosis.

Eighty winter-tender clones selected for good performance against pea aphid in a large cage at Logandale, Nev., were given several tests in stem-cages at Tucson, Ariz., to isolate the clones with the highest antibiosis to this insect. Thirty-eight were selected for further study.

At Poznan, Poland, under a P. L. 480 project, two Nevada selections of alfalfa, Medicago sativa, (Nev. 416 x 1-113 and Nev. 552 x 609) were compared for pea aphid resistance with the Polish alfalfa variety Miechowska and also with M. lupulina. None of the aphids feeding on M. lupulina survived more than 5 days. At the end of a two week period 520 aphids were alive on the Polish variety, Miechowska, compared with 120 and 99 on Nevada varieties 552 x 609 and 416 x 1-113, respectively.

6. Meadow Spittlebug. A method was developed at University Park, Pa., for screening alfalfa in the greenhouse during the winter months for resistance to the meadow spittlebug, Philaenus spumarius. The procedure consisted of planting oats in rows in greenhouse flats in April, caging field-collected adult spittlebugs for oviposition on the oat stubble in these flats in September and October, and storing them at 36 to 40° F until mid-December. Alfalfas to be screened were established in 1-inch peat pots in September. After mid-December flats were removed from cold storage and the potted alfalfas were planted between the rows of stubble. Flats with plants were held at 65 to 70° F in the greenhouse for 7 weeks and were then appraised for spittle masses and nymphs per plant. Masses ranged from 0 to 10, and nymphs from 0 to 26 per plant. Screening from January to May will allow more time for breeding studies with the more promising insect-resistant plant material.



7. Lygus Bugs. After two years of field screening, 38 alfalfa plants were selected for antibiosis cage testing in the greenhouse at Mesa, Ariz. Preliminary studies showed that 5 plants held promise for antibiosis type of resistance after 2 separate cage tests using third instar nymphs. Resistance was based on percentage of nymphal mortality using 5 nymphs per caged flower bud.

Research is being initiated at Manhattan, Kans., to develop techniques and screen alfalfa plants for resistance to lygus bugs and other mirid species under a grant awarded to Kansas State University of Agriculture and Applied Science.

8. Alfalfa Seed Chalcid. In studies at Mesa, Ariz., adults of the alfalfa seed chalcid, caged on the flowers, leaves, stems, and racemes of resistant and susceptible alfalfa plants, lived longest, 8.23 days, on the flowers. The average longevity on leaves was 6.51 days; stems, 4.61 days; and racemes, 4.42 days. There was no appreciable difference in longevity between resistant and susceptible plants. Longevity appeared to be influenced by rate of plant transpiration which was highest on the leaves.

9. Sweetclover Weevil. While an excellent source of resistance to the sweetclover weevil has been found in Melilotus infesta, this species does not readily cross with the commercial species. During the past winter evaluations were made on 24 accessions of M. dentata which, with the use of special techniques, will cross with M. alba. Three appeared to have intermediate levels of resistance and plants were selected from them for further study. Studies involving grafts between sweetclover weevil resistant (M. infesta) and susceptible (M. officinalis var. Goldtop) plant material show that there is little movement of the resistance factor across the graft union.

A grant jointly with the Crops Research Division was recently awarded to the Nebraska Agricultural Experiment Station to study the nature of the resistance of sweetclovers to the sweetclover weevil.

10. The Spittlebug, Prosapia bicincta. At Tifton, Ga., 81 bermudagrass hybrids were screened for resistance to damage by spittlebugs. Adults of both sexes were caged on the different grasses growing in 2-inch pots. The grass was observed daily for symptoms. None of the grasses showed resistance.

#### H. Insect Vectors of Diseases

1. Clover Viruses. At Poznan, Poland, under a P.L. 480 project bean yellow mosaic virus and red clover vein mosaic virus were simultaneously transmitted from crimson clover plants infected with both viruses to healthy crimson clover plants, by the green peach aphid, Myzus persicae, and the pea aphid, Acyrthosiphon pisum. Both viruses were successfully transmitted to 50% of the test plants by the green peach aphid and to 40% by the pea aphid. Bean yellow mosaic virus alone was transmitted to 33.3 and 38.2% of the plants by the green peach aphid and the pea aphid, respectively. Red clover vein mosaic

virus was transmitted to 13.4 and 18.8% of the plants by these aphids. No symptoms appeared in 3.3% of the crimson clover plants fed on by infected green peach aphids and 3.0% fed on by the pea aphid.

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PRODUCTION, HARVESTING AND FARM STORAGE STRUCTURES,  
EQUIPMENT AND METHODS  
Agricultural Engineering Research Division, ARS

Problem. Better methods, techniques, equipment and structures for use on the farm for producing, harvesting, storing, and the initial preparation of grain, seed and forage crops for market are needed to increase efficiency in the use of labor and equipment, preserve quality and prevent spoilage and damage from mechanical handling and insects. Electro-magnetic energy has many established farm uses but research indicates other highly useful potential capabilities in farm production, such as attracting and killing insects and improving the germination of seeds. There is considerable present need for precise seedbed requirements for various crops in different areas of the country. The exact best placement for starter fertilizer is also unknown for a number of crops under different climatic and soil conditions. Every method to control or eradicate plant diseases, weeds, and insects requires some type of equipment. Effectiveness of the equipment necessary may be essential to the success of the method which is attempted or recommended. The cost of harvesting and farm handling of forage, grain and seed crops is the major expense of production. The supply and adequacy of manpower for these operations are becoming progressively less satisfactory, thus requiring additional research in this area. While considerable information has already been obtained for the development of processes such as drying and separation, basic and more precise information must be developed for these and other processes in order to achieve further progress. New trends in storage structures also require additional research in design and use.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long term program involving agricultural engineers, statisticians, physicists, and architects engaged in both basic and applied research in this area. Cooperation is maintained with experiment stations, the farm equipment industry and other scientific disciplines in the Department. The total effort involves 25.7 professional man-years. Of this number, .8 are devoted to the use of electrical energy, 1.3 to fertilizer placement distribution and transplanting equipment, 2.4 to equipment for establishment of forages, 2.0 to weed control in corn and soybeans, 4.9 to insect control in grain, 1.5 to forage harvesting equipment, 1.5 to forage seed harvesting equipment, 5.4 to grain drying, 1.5 to forage processing, 2.5 to seed cleaning and 1.9 to crop storage structures. Research is carried on at Beltsville, Md., Athens, Experiment and Tifton, Ga., Ames, Iowa, Lincoln, Nebr., Corvallis, Ore., Lafayette, Ind., Bushland, Tex., and Manhattan, Kans.

## PROGRAM OF STATE EXPERIMENT STATIONS

Many of the State agricultural experiment stations are engaged in both fundamental and applied research dealing with the development of new principles and the application of currently available knowledge to the problems concerned in soil-machine relationships in order to increase efficiency in crop production. These studies are concerned in the broadest sense with the development of theories, special devices, and laboratory and field tests to determine ways in which tractive and transport equipment, tillage tools and systems for their use might be improved.

Problems concerned with planting of the many sizes and shapes of seed together with the introduction of fertilizers are under attack by many of the State agricultural experiment stations. These studies are concerned with the development of new principles that can be used to meter and place seed which could lead to planter improvement. Similar investigations are in progress to develop satisfactory metering and placement devices for application of liquid as well as solid fertilizers. In both instances the principal objective is to provide the best possible means of seed and fertilizer placement which will assure healthy plant emergence with vigorous growth to maturity.

Both basic and applied research investigations which have been designed to discover and develop methods, techniques, and equipment for control of the many pests that attack grain and forage crops are in progress at the several agricultural experiment stations.

Most of the State agricultural experiment stations are engaged in some aspect of basic or applied research which is concerned with improving machines and methods for efficient harvesting and farm handling of grain and forage crops. Detailed investigations are in progress to develop reliable mechanical harvesting and handling equipment as well as ways in which improvements might be made in crop production systems to increase yields, product quality and overall efficiency.

Many freshly harvested agricultural crops must be subjected to early treatment in order that they may retain as much as possible of their original qualities. The State agricultural experiment stations are involved in both basic and applied research studies which have as their broad objectives the development of improved methods, equipment and techniques for preparation and processing of farm crops to preserve quality and prevent spoiling while in storage.

The complicated problems associated with providing protection to the products of agricultural production as well as the machines, equipment and service facilities which are required for such production has necessitated a continuing program of research at the State agricultural experiment stations. The current program is concerned with conditioning and storages for high moisture grains; structural characteristics, wall pressures,

design and construction of silos; controlled atmosphere storages and construction methods; and designs and construction of plant growth chambers and plastic greenhouses.

Several of the States are engaged in programs of basic and applied research on the possible use of some of the various forms of electrical and physical energies as a means for improvement of the potential capabilities in farm production. Investigations in progress include the evaluation of the use of radiofrequency energy for treatment of grains to destroy insect infestation and treatment of seeds to improve their germination characteristics.

The agricultural experiment stations of many of the States have research under way whose major objectives involve the obtaining of information on the uses to be made of electrical energy to reduce labor, increase production and improve family living conditions. In the design of these studies provision has been made to develop and investigate new equipment and explore the possibilities for new uses for electricity on the farm. Many of the projects are concerned with the varied problems of chore labor mechanization and an expansion of the use of electricity for ventilating, heating, lighting and cooling under the various production enterprises of today's farming operations. Development and testing of prototype specialized equipment for product collection, processing, packaging, and transport, as well as crop storage, loading devices, are a part of the overall program of investigations.

The State agricultural experiment stations are engaged in extensive basic and applied research to extend the advantages of controlled environment to all phases of agriculture in order to obtain maximum economic growth, production, product preservation and product quality. Studies of the possibilities for use of solar energy as well as electric energy to achieve the broad scale objectives are a part of the total program. Among the several investigations involved in these programs are determination of the effects that soil, light and atmospheric conditions on plants; and temperature, humidity and gases on stored products. Special attention is being given to development of means for collection, storage and use of solar energy for crop conditioning.

Much of this research is cooperative with the Department.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAM

##### A. Systems of Equipment for Conservation Farming

Studies were continued in Iowa on the use of herbicides to reduce or eliminate seedbed preparation. Overall spray applications of atrazine, simazine, and 2,4-D sprays were made in early spring on unplowed and spring plowed cornstalk ground. Secondary tillage treatments included disking and



harrowing, disking, strip tillage with cultivator sweeps, strip tillage with a rototiller, and no secondary tillage. The control or check treatment to which all others were compared was conventional moldboard plowing, disking, and spike tooth harrowing. The yields, stand, and weed weight data collected show that on spring plowed land most of the secondary tillage was unnecessary. Atrazine was the most successful chemical used. On unplowed cornstalk land, yields were lower than normal because of the high moisture conditions that persisted during the early growing season. None of the chemicals performed well enough to eliminate all mechanical cultivations. However, where atrazine was used, only one cultivation was required. Satisfactory stands were usually not obtained where no tillage was performed. A single disking or strip tillage over the old corn row to a depth of not more than two inches appears to be adequate to obtain satisfactory stands. These results indicate that corn can be successfully grown in Central Iowa with little or no seedbed preparation if adequate stands and weed control can be obtained.

Studies were continued on equipment and methods for preparing seedbeds and controlling weeds on ridge planted corn. Through the use of pre-plant applications of herbicides on contoured ridges that had been formed 12 years ago, it was possible to obtain almost complete control of runoff and erosion and to maintain a high level of stands and yields. Comparisons of growing continuous corn on ridges that were constructed each year and continuous corn grown in the conventional manner show that the two systems are equivalent where similar stand levels and weed control were obtained. Chemical weed control was somewhat more consistent than mechanical methods with the ridge planting system.

Studies were conducted on the effect of moldboard plows, disk harrows, spike tooth harrows on the soil density, clod size, and surface roughness. Results showed that the moldboard plow decreases the density of the soil plow layer by 25 to 35 percent. Subsequent disking and harrowing increases the soil density. The degree of compaction appears to depend on soil type as well as on moisture content. An examination of clod sizes ranging from 1/8 in. to three inches in 1/8-in. increments showed that the plow produced the largest clods and that subsequent operations caused only a minor change in the clod size distribution. Changes in surface roughness before and after tillage showed that the plow resulted in the roughest soil surface and that subsequent operations tended to smooth or level the soil surface. It appears that one disking and one harrowing would be adequate but that three diskings would usually result in a slightly smoother soil surface.

Studies with model moldboard plows were conducted to develop a functional relationship between measurable variables that would be useful for pre-dicting the forces on full-size moldboard plows working in various soil types. These studies were conducted in small soil bins where the soil variables could be controlled and the forces on the tools could be accurately measured. Measurements were made of the soil strength properties

and the draft forces on model moldboard plows operating at different speeds through various soils. The data were used to develop a functional relationship between dimensionless draft force terms and other dimensionless terms made up of the measurable variables. A functional relationship was developed for mathematically predicting draft forces on moldboard plows. This relationship was found to be valid for model plows of a given configuration operating in soils exhibiting a rather wide range of soil strength values. In these model studies, real soils were used, and apparent cohesion was a dominant soil strength property. Test results indicate that the selection of apparent cohesion and the tangent of the angle of shear resistance are pertinent soil strength properties. Although it was not possible to determine the effects of apparent adhesion and the coefficient of soil to metal friction, it is believed that these factors are also important.

#### B. Fertilizer Placement and Distribution Equipment

Eighteen experiments on the establishment of field crops and vegetables were put in cooperatively with various State, Federal and commercial research units. Some typical results or trends shown of the field experiments of field crops or vegetables are: Winter wheat yields in cooperative field experiments with Michigan averaged 51 bu/acre, while check plots with conventional openers with fertilizer in contact with seed averaged 35 bu/acre. These findings conform to the results in two other states (as reported last year) and the separate seed-fertilizer opener developed by the project for close drilled seed has proved quite successful on the special machines for research. One of the major implement companies has issued a seed-fertilizer opener patterned after our development. Although it is quite successful for experimental and slow speeds (up to 2 mph) for field use, there is need for further development to meet the present day high speed planting requirements (up to 5 mph).

#### C. Equipment for Establishment of Forages

1. Sixteen experiments on the establishment and production of pastures and haylands were put in cooperatively with State and Federal research units. Some typical results that show response or definite trends from new planting or application methods in the various studies are: In a first year experiment with methods of establishing Coastal bermudagrass in the southeast, superior stands resulted when a small amount of water was placed with each sprig that had half of the fertilizer side placed in two continuous bands at time of sprigging. The first six weeks after sprigging were high in temperature, and no precipitation. The stands and final ground coverage were better than irrigated treatments and this treatment with no herbicides equalled the other half of the plot treated with herbicides. This was a cooperative study with the AE and Agronomy Departments of the University of Georgia, and the SWC station at



Watkinsville. In interseeding Birdsfoot trefoil in bluegrass sod in Maryland, experimental seed-fertilizer openers were able to establish a six-month stand which average about 25 plants per running foot. The plots interseeded by commercial seed-fertilizer sod openers had a zero stand after six months.

Cooperative studies on planting methods and means of applying fertilizer on forage crops in the southwest area were conducted in four states (Texas, New Mexico, Oklahoma and Louisiana). The studies involved various cultural methods on grass seeding, interseeding grass on rangeland and on abandoned cropland, and sand dune stabilization. In regard to grass establishment as affected by soil compaction, forage yields went down as the soil strength went up. Better seedling emergence and establishment were obtained by late seedings (June). After two seasons of interseeding side-oats grama grass in abandoned farmland, shallow seeding late in the spring gave highest survival of seedlings. One half inch seeding depth was superior to one inch depth (1963 season only). May and June seedings gave highest seedling survival (1963 & 1964). In sand dune stabilization (Oklahoma) the fertilization of existing plant species appears to be promising in solving sand dune problems within reasonable economical limits. Vegetative mulches to establish vegetative cover indicate superiority to chemical mulches, but both are prohibitive (costwise) at present day values. Growing mulch in place (sorghums or small grains) so far has not been feasible.

2. A contract was signed in June, 1964, whereby the New Mexico Agricultural Experiment Station is to conduct 3-1/2 years of research for the Department on the design and development of range seeding equipment for use with brush eradication equipment in the arid Southwest. The primary objective of the research is to modify existing equipment or to develop equipment that will accomplish the following four operations in one pass over the land: eradicate undesirable shrubs, provide a firm seedbed, plant seed effectively, and windrow plowed-up brush over the planted strip.

Three major types of equipment have been investigated: a rootplow, a seeder to firm the seedbed behind the rootplow and effectively plant grass seeds, and windrowing equipment to place plowed-up brush over the planted strip to give shade and wind protection to emerging plants. A rootplow with an 8-ft. horizontal blade, commercially available, operating at a depth of about 10 in., proved effective in eradicating creosotebush and tarbush. No modification of the rootplow was necessary. A range seeder, also commercially available, was attached behind the rootplow and seeded four rows spaced one foot apart, making a 3-ft. wide seeded strip. After some minor modifications, the range seeder satisfactorily provided a firm seedbed and planted seed effectively.

Several types of equipment were tried for windrowing the plowed-up brush over the planted seed, but without entire success. When a grader blade was operated at from about 40° to 60° with respect to the rootplow blade, portions of the brush became caught between the grader blade and the ground.



When a modified blade, 14-ft. long, was attached behind the rootplow, no practicable angle could be found at which the brush would move satisfactorily across the face of the blade. A hay windrower attachment, normally used behind a 7-ft. mower cutter bar, was tried for moving the brush over the seeded strip, but which tended to catch and drag the brush instead of moving it laterally over the seeded row. A side delivery hay rake with rotating tines was tested back of the rootplow for moving the brush over the seeded row. Observation of this rake indicated that the principle of operation may be suitable if the rake wheels are shielded. Additional studies of brush windrowing equipment will be made.

#### D. Insect Control in Grain

1. Insecticide granules were applied for control of Northern corn root worm in cooperation with the Ohio Station, with equipment developed or adapted for small plot treatment. A fluted-type feeder gave extremely accurate application rates of a large number of different granular materials used. In one experiment using 12 different insecticides only one, heptachlor, was significantly better than the untreated check, yielding 18 bushels of corn per acre more than the check. No significant differences were found among the roots damaged and the number of plants that were lodged.

In investigations in cooperation with the Iowa Station, the major emphasis of the program was the screening of new insecticides, development of systemic insecticide control, and development of combined control of the European corn borer and corn rootworms. The results of these studies indicate that several experimental compounds may be as effective for borer control as presently recommended compounds. American Cyanamid 47470 was shown to be the most effective compound for systemic control of corn borers, however, Niagara 10242 demonstrated some activity. Experiments in 1964 show it is possible to combine the control of first-generation corn borers and corn rootworm larvae with one application of insecticide.

2. Ultrasonics are being investigated in Georgia as a possible means of controlling insects on southern grain crops without chemicals. Field work has been concentrated on repelling corn earworm moths, while laboratory work has been devoted primarily to destruction of eggs and larvae. Ultrasonics used in the laboratory did not reduce armyworm or earworm egg viability within an exposure time of five minutes or less. However, ultrasonics killed the first instar larvae of either the armyworm or earworm in less than 15 seconds.

Seeds of corn were oriented in Georgia at the time of planting to determine if the foliage and ears could be oriented and, if so, would the orientation effect insecticidal application for insect control. These tests did not show any orientation of the ears or foliage, with the ears growing about the same in each direction. Yields were slightly higher from the oriented rows. Insect damage was not significantly different between the oriented and unoriented rows, receiving the same insecticide treatments.

3. DDT was further evaluated in Georgia for its effectiveness in controlling corn earworms in sweet corn. Three plots were treated by recommended procedures with 2 lbs.DDT/A. One plot was treated every day, another plot was treated every second day, and the other plot was treated every third day. The plot receiving a treatment every day resulted in 66 percent worm free ears. The plot treated every second and third day had 50 and 53 percent worm free ears, respectively. Effective earworm control was not obtained.

A study was made in Georgia for determining residues and insect control in sweet corn when applying an insecticide in the form of a dust, liquid and granule. The dust was applied through nozzles positively charged with 13,200 volts of electricity, negatively charged at the same voltage, and uncharged. The emulsifiable concentrate liquid was applied by spraying and with a brush applicator. Spraying gave the best insect control, while the brush applicator was next. Granular form was decidedly the poorest. The brush applied more insecticide on the target area (ear tips) than the spray, and used 1.6 lbs. DDT/A against 2 lbs. DDT/A applied with the spray. Dust from positively charged nozzles gave the highest dust deposition.

#### E. Weed Control in Corn

1. Several methods of applying preemergence chemicals for weed control in corn were used in investigations cooperative with the Iowa Station. Mixing granular and liquid formulations of atrazine, 2,4-D, and Radox T into the soil at planting time with rotary hoes, drag harrows or wire wheels did not improve the effectiveness of the compounds. With atrazine and simazine liquid preemergence applications gave better weed control than granular formulations. Granular and liquid formulations of 2,4-D and Radox T applied at planting time were equally effective. None of the preemergence treatments gave full season weed control, and one additional mechanical cultivation was required for band treatments.

Studies on equipment, techniques, and chemicals for directional postemergence sprays in corn were continued in Iowa. Results were erratic but did show that this practice has possibilities as a "salvage" operation. Studies on weed control practices for narrow-row corn indicate the need for little or no change in mechanical or chemical methods. Earlier soil shading with narrow rows resulted in improved weed control. Timely mechanical cultivations gave better weed control in narrow-row corn than most chemicals.

Early spring applications of atrazine and simazine in Iowa on plowed soil successfully controlled weeds in corn throughout the growing season without secondary tillage or cultivations. Similar applications on unplowed cornstalk ground were nearly as successful but required one mechanical cultivation.

Field studies were made in cooperation with the Missouri Station to evaluate different methods for directed applications of dalapon (1 & 2 lbs./A) for weed control in corn. Directed applications were made that varied from no leaf protection to maximum leaf protection provided by tying the leaves to the corn stalk. Three different applicators (a special shield leaf lifter, a wire leaf lifter and a special directed nozzle) were used in the study. Applications were made to corn under weed-free conditions so corn damage from contact with the dalapon could be evaluated. No significant corn damage was noted for any of the treatments. This is the first year of a three-year study where no damage occurred. This is thought to be due to the absence of rainfall after application this year. Additional studies will be made under both weed-free and weedy conditions. A field study was made to determine if incorporation of amiben (1, 2, 3 and 6 lbs./A) would allow its use for weed control in corn. However, incorporation did not reduce the damage to the corn. All treatments resulted in lower corn yield than for the cultivated check treatments.

Field studies were continued in Missouri to determine the effectiveness of 8, 10, 12, 14 and 16-inch band applications, sprayed over the rows, of 2,4-D, simazine and atrazine for weed control in corn. In each case, the amount of active ingredient for the area actually covered was the same, resulting in a saving of chemical for the narrower bands. The middles between the rows received one cultivation and it was more difficult to cultivate the plots that were treated with the narrower bands of spray. No significant difference in corn yield was noted for any of the band applications.

The direction of movement of the herbicides in soil was studied during the band applications in corn in Missouri. In the crop year 1963, under dry soil conditions, weed control was effective over a wider area than the applied band. In 1964, control was limited to the actual area under the band. This would denote that there is greater lateral movement of the herbicide under dry soil conditions, however, this study will be continued in order to establish conclusive results.

#### F. Forage Harvesting Equipment

1. In Georgia, the effect of vapor barriers, between the ground and the Coastal bermudagrass on drying rate, was insignificant when drying conditions were good and the ground was dry when the hay was cut. Under poorer drying conditions and with damp ground, however, hay placed on a vapor barrier dried more rapidly. Although hay on the vapor barrier dried more rapidly, it did not reach safe baling moisture with only 1-day's drying. Since hay without a vapor barrier could also be baled the following day, the use of a vapor barrier does not appear feasible.



2. Low moisture alfalfa silage. A rapid, accurate method for determining losses in harvesting hay-crop silage was developed. The use of a windrower and pickup chopper gave dry matter yields similar to direct-cut chopping. Conventional mowing and raking before pickup chopping resulted in lower harvested yields, regardless of the moisture content of forage during raking. The dry matter bulk densities of hay-crop silages stored in tower silos were little affected by lengths of cut or by original moisture contents. The average density of low-moisture silage that had been chopped with a machine set at three-fourths inch theoretical cut was only slightly less than that chopped more finely by a machine set at one-fourth inch cut. Also, the dry matter in low-moisture silage was only slightly less dense than that of high-moisture, direct-cut silage.

#### G. Forage Seed Harvesting Equipment

1. Optimum moisture content for seed harvesting. At Corvallis, Oregon, research on the time of harvest was continued on bluegrass and orchardgrass to determine the optimum stage of maturity at which to cut the crop in order to obtain the maximum quantity of pure live seed. In bluegrass harvesting, the 25 percent seed moisture seems to be the optimum time to windrow the crop, but this will require more research to be conclusive. In orchardgrass harvesting, when the crop was cut at 44 percent seed moisture, the maximum quantity of pure live seed was obtained. The 1964 yield was 776 pounds per acre when cut at 44 percent moisture, as compared to 304 pounds per acre when cut at 18 percent seed moisture. By cutting the crop at 44 percent seed moisture, the increase in seed harvested on the present acreage would far exceed the 4.5 million pounds imported each year. A simple inexpensive moisture meter that uses the exhaust heat from a tractor, truck, or any other internal combustion engine was developed to determine quickly and easily the seed moisture in the field. The research on six crops indicates that seed moisture is a reliable method of determining seed maturity.

2. Development on components for cutting, picking up, threshing, and cleaning field seed crops. A rotary cutter was altered for the purpose of cutting and removing seed crops from the field. When harvesting crimson clover with the altered machine, 94 percent of the crop was cut and delivered to a wagon with very little seed damage. The unit was less effective in harvesting row crops, as only 50 percent of the production of bluegrass, orchardgrass, and birdsfoot trefoil planted in rows was saved. However, the overall test results indicate that the rotary cutter can be a satisfactory replacement for the cutter bar on a combine for a number of seed crops.

## H. Seed Cleaning

1. Seed cleaning research applied to specific problem mixtures. Processing research was conducted on 30 seed mixtures furnished by the seed industry in order to develop techniques, procedures, and equipment to make the separations that would salvage the crop seed.

The technique of microscopically measuring the length, width, and thickness of the crop seed and its contaminants, has proven very effective. This technique was used to select a 6 x 26 slotted-hole screen which proved most successful for removing rattail fescue from creeping red fescue. In this case, the purity was raised from 85 to 99.6 percent.

Data obtained by this technique were used to design a special indent cylinder that would remove six of seven kinds of weed seeds commonly found in bentgrass. Oregon's annual production of bentgrass is valued at more than \$2,250,000. The special indent cylinder, designed from the research data, with indent pockets measuring 0.035 inch in diameter and 0.012 inch in depth was used to lift red sandspurry from a 20,000 pound lot of Penncross bentgrass at the rate of 50 pounds per hour, thereby reducing the weed count from 2.5 percent to .04 of 1 percent. Removal of the weed seed increased the value of the seed \$.85 per pound, or \$17,000 for this one seed lot. The same procedure was used to design a special indent cylinder with pockets 0.062 inch in diameter and 0.019 inch in depth to remove pigweed from alfalfa. This development can save the seed industry several million dollars annually.

Another development is a machine that separates seeds by differences in resiliency. This machine reduced the percentage of ryegrass from 5 to 0.5 percent by "bouncing" ryegrass out of orchardgrass.

The surface-textured deck vibrator separator research tests have resulted in several additional difficult separations being made. A 500-deck machine of this type was constructed by a processing plant in accordance with specifications furnished by the project's personnel. The machine is now being used in a commercial seed processing plant to convert ladino clover screenings into salable seed at a rate of 200 pounds per hour. It is estimated that \$19,000 worth of seed will be saved in a single season at the one processing plant.

2. Development of centrifugal-pneumatic seed separator. Considerable progress was made in the development of a pneumatically cleared vertical rotating screen. Preliminary tests indicate that when operated at 300 r.p.m., which generates a centrifugal force 10 times the pull of gravity, the unit will make a more efficient separation at 3 times the capacity of that of the conventional flat vibrating screen of the same surface area. Materials run

in the machine thus far have been wheat, crimson clover, fine fescue, sunflower seed, and sawdust. The unit is a revolutionary step in screening, and has already attracted the attention of machinery manufacturers; for example, the farm machinery companies are interested in the unit for use on combines, the air-screen manufacturers are interested in it for seed processing, and other manufacturers such as mining and building contractors are interested in it for their numerous screening and sizing operations. The manufacturers in all three areas have obtained detailed information on the machine and have constructed test units. When the centrifugal screen machine is fully developed, it is estimated that it will mean a saving of several million dollars annually to the above industries.

3. Modification of seed-length separators. The scientific approach of microscopically measuring dimensions of seeds in problem seed mixtures resulted in the development of a number of special indent cylinders that make specific separations.

The seed industry is already using special cylinders to separate red sand-spurry, yellow cress, toad rush, mouse-ear, and other weed seeds from the bentgrass, and to separate pigweed from alfalfa.

The amount of seed saved is estimated to add 2 million dollars annually to the farmers' revenue.

#### I. Grain Drying

1. At Ames, Iowa, studies are underway for isolating the factors that enter into rational design of drying equipment and for developing quantitative descriptions of their relation to economic design. Measurements of the rate of moisture loss in thin layers of kernels are being made to determine the separate effects of air temperature, air humidity, air velocity, initial grain moisture, initial grain temperature, and other grain properties on moisture movement. Also observations are being made on the effects of contraction and expansion of grain as its moisture content changes. This year's tests have included soybeans as well as corn. The data from corn show a response to drying conditions that are similar to, but more complicated than, that found for grain sorghum kernels. This difference is presumed to be due to the nonspherical shape of the corn kernel. Tests with relative humidity as near to zero as possible indicate that some of the moisture that might be removed from a kernel at high temperature (for example, 200° F.) cannot be removed even with a very extended period of exposure at zero humidity when the temperature is lower (for example, 120° F.). This suggests that a portion of what may be defined as "moisture" at one temperature may become a part of the "dry matter" at another temperature.



2. Studies of the drying zone in mechanical grain driers were continued through use of laboratory model drying bins. The drying zone refers to that part of the grain in a drying bin in which drying is actually taking place at any given time. The zone moves through the grain but, in general at any specific time during drying, part of the grain is drying, part is already dry, and part is still at its initial moisture. The temperature and moisture gradients in the drying zone determine the efficiency and the required drying time in any drier. Accurate definition of the drying zone in terms of grain moisture, air temperature and humidity, rate of airflow, etc., would permit rational design of drying equipment. While much progress has been made in relating these factors to the drying zone, drier design still remains an art without the support of adequate design data.

3. Effects of mechanical damage to field-shelled corn. Attempts have been made to evaluate the damage to kernels in samples from various shelling treatments by immediate inspection. The severity of damage by various machines is difficult to assess and little progress has been made. The most promising method so far appears to be treating the samples with fast green dye so the skin breaks become more apparent. In a field test with different types of shellers and various row spacings no preference could be detected for any machine or method. Tests of flaming corn ears to partially sterilize the grain and to temporarily harden the kernel surfaces prior to shelling did not improve the condition of the corn after shelling.

4. The time limitation on deep bed or in-storage grain drying systems is dictated by grain deterioration which is caused primarily by the growth of molds and bacteria. Of secondary importance may be the respiration or growth of the seed itself. The factors which influence the rate of growth of the microflora are grain moisture, temperature, and the amount of physical damage of the grain. It is the purpose of this study to evaluate the influence of these factors on the rate of growth of the microflora and subsequently the rate of deterioration.

Generally the tests show a tendency for the CO<sub>2</sub> production rate to increase with time, but for this tendency to be less pronounced at high temperatures. In one series the rate was almost constant at 75° F. In another, this did not occur below 110° F. Further research will be required to establish relationship between rate of CO<sub>2</sub> production and different combinations of time and temperature.

The effect of mechanical damage on permissible drying time reported previously has been further confirmed. However, damage inflicted by razor cuts had far less effect than that caused by field shelling. Whether this difference is due to the inoculation provided in the shelling mechanism or whether a simple skin rupture is only a small part of the mechanical damage caused by the machine remains to be determined.

## J. Forage Processing

1. Tests with a stationary wafering machine showed that a wide variety of products could be wafered or cubed. Coastal bermudagrass, peanut hay, gin trash and corn cobs and husks were all wafered without grinding. Due largely to an inefficient feeding system, the production rate was low and the power requirements high. With modifications, it appears that this process might be feasible.

Tests indicate that dehydrated and pelleted small grain forage retains the milk stimulant present in the fresh forage. Although the material pellets easily, the energy costs for processing are just about double that for processing Coastal bermudagrass. The increase in energy is due to the large amount of water that must be removed for each ton of dry matter.

A 3-year study of systems of utilization of Coastal bermudagrass showed that dehydrated and pelleted hay gave the best animal performance. Following, in order of decreasing rate of gain, were dehydrated hay, continuous grazing, rotational grazing, strip grazing and green chop. The calculated gain per acre for the two processed treatments was about 300 pounds per acre more than for continuous grazing. The processing energy costs were \$39.02 per acre for dehydrating and pelleting and \$34.00 per acre for dehydrating only. This concludes these studies. Next year a study of the processing costs and relative value, measured by animal gains, for pellets made from dehydrated, partially field-cured and field-cured hay will be undertaken. Tests will be made using 4- and 6-week old grass.

Effect of unit processes involved in dehydrating and pelleting on the economically important constituents of Coastal bermudagrass and Pearl millet. Preliminary tests were run to determine the minimum sample size for testing the effects of each of the processes, dehydrating, grinding and pelleting, and the effect of additives on these constituents, and on the feasibility of dehydrating and pelleting millet which was older than 4 weeks. During the coming year these tests will be run and information developed for linear programming for use by the feed processing industry.

2. Chopping or grinding hay prior to wafering resulted in wafered products having different physical and nutritional qualities. Baled hay was flail chopped, ground through a large screen or through a small screen, and then wafered in a stationary system. The most dense wafered product with the least amount of fines was made from the hay having the smallest size particles. Sheep ate this product in the greatest quantities.

Physical qualities of samples of wafered hay were related. Higher bulk densities of samples of wafered hay were associated with a lower percentage of fines (material not in wafers). However, the bulk density of the fines tended to be higher for those samples with higher overall bulk densities. Within the limits experienced, the percentage and bulk density of fines had little effect on the overall bulk density.



## K. Crop Storage Structures

1. Silo design criteria. Determination of forage density in normal storage conditions, using a radioisotope, continued at Beltsville, Maryland. This year's work was aimed at finding effects of length of chopped alfalfa on stored silage density. Measurements were more precise than in previous years. Chopped alfalfa in chop lengths averaging 1.34 and 0.80 inches was loaded in a silo with the two chop lengths in alternate layers. The long chopped layers averaged 95% and 91% as dense as the short chopped layers at 1.8 and 21 days after loading, respectively. There were indications that settling proceeded unevenly around the silo circumference. It is planned to determine density in a silo loaded by typical farm procedure next year.

At Athens, Georgia, research continued on measurements of the gaseous transfer rate of silo construction materials. Effort during the year was on measuring the flow of oxygen through concrete silo walls. Flow rates through basic silo staves were determined under conditions of equal total pressure; hence, flow was caused by a pressure gradient across the concrete sample. Treatments in the study involved coatings of plaster, latex paint, and a special polystyrene paint. Results indicate that oxygen diffusion through silo walls could be greatly reduced by the application of an inexpensive coating which is relatively impervious to oxygen flow. This project was actively conducted for only nine months this report year. The work is being phased out at this location and no additional activity is planned.

On studies of pressures in large tower silos, at East Lansing, Michigan, no reportable progress was made.

2. Heavily wilted silage storage. In studies at Beltsville, Maryland, dry matter density of heavily wilted alfalfa in tower silos was 15% less at 64% dry matter than at 44%, which represents a decrease of silo capacity and more difficulty in sealing with low moisture silage. This, plus the chance of weather damage during heavy wilting, is an important deterrent to using very dry silage. Visible spoilage in storage was found to have no simple relation to total storage losses. Feed dry matter recoveries from 3 farm size silos were 89%, 90%, 91% from respective average dry matters of 44%, 58%, 64%, in contrast to a similar test last year from which recoveries were inversely related to dryness. Further trials will be run to reconcile these findings. A trial of making heavily wilted orchardgrass silage in a bunker during adverse weather produced silage which heated for 25 days and yielded only 72% recovery from storage, which shows the disadvantage of attempting heavy wilting during poor drying weather. Plans for next year are a comparison of 2 chop lengths of wilted alfalfa for storage efficiency and handling problems.



3. Bunker silo pressures. No reportable progress was made on the study of pressures on bunker silo walls at Beltsville, Maryland, due to the scarcity of forage at Beltsville. Work is expected to continue whenever full loads of critical crops are available for the test silos.

4. Hay wafer storage. No work was done on the hay wafer storage project at Beltsville, Maryland, during the reporting year. Consideration is being given to resuming this work at a later date.

5. High moisture shelled corn storage. At Ames, Iowa, no additional field work was done on storage of high moisture shelled corn. Data obtained in previous studies were analyzed for the effects of gaseous (oxygen-carbon dioxide) environment on rate of growth of microorganisms such as molds, bacteria, and yeasts. This rate of growth is a function of the moisture content, temperature, and available food or physical damage of the kernel coat and also may be considered as an oxidation resulting in production of carbon dioxide. Since the evolution of carbon dioxide can be readily measured, it can be used as an index of deterioration.

Molds, coliform bacteria, mesophilic aerobic bacteria and yeasts had been enumerated at intervals during ensiling. The numbers of aerobic bacteria were similar in structures containing different concentrations of gas and held at different temperatures. Coliform bacteria could not be detected after 10 days of ensiling. Mold numbers were relatively low, but were important in the deterioration of corn at the surface of the silos. Yeast and bacterial numbers increased rapidly following an initial aeration period, but no increase was observed after a second aeration series. Lack of a growth response to the second aeration series is believed related to the depletion of an assimilable carbon source. Yeasts were preeminent in proper preservation of high moisture corn.

#### L. Electric Traps for Grain Insects

Cooperative studies with Purdue University were conducted to determine the responses of cereal leaf beetles to electromagnetic radiations and to determine whether light traps could be used to survey for this pest.

In laboratory studies, adult beetles approached both green and blacklight attractant lamps placed at the end of an 8- by 4- by 4-ft. chamber. None were observed in flight. An additional test indicated that cereal leaf beetles were not attracted to a 60-watt incandescent lamp.

Temperature was a critical factor influencing flight in both field and laboratory investigations. At 70° F. the insects crawled or hopped; at 75° and 80° they readily flew. In laboratory tests at 80°, 74 percent of the

beetles released were captured in traps using one 15-watt blacklight or one 15-watt green lamp as the attractant.

Field investigations were continued to determine the feasibility of using light traps to survey for cereal leaf beetles. Traps were installed in a heavily infested area in southern Michigan and in a lightly infested area in northern Indiana. Trials were inconclusive in Michigan because an insecticide application drifted several miles into the test area, killing most of the adult cereal leaf beetles and nullifying the endeavor. Only three beetles were caught in northern Indiana, with very few observed in the nearby area. After 2 years' investigation, results are still inconclusive but indicate that light traps will not be useful as survey tools for cereal leaf beetles until the insect pest spreads to a warmer climate.

European corn borers caused an estimated 50-million-bushel corn loss in Iowa in 1964, the greatest loss in 5 years. Light traps are useful in studies and in the control of corn borers by determining the time of emergence, the dates of maximum flight activity, the comparative activity from year to year, and the timing of insecticide applications. Light traps have been operated near Ames since 1952 in cooperation with the USDA European Corn Borer Investigations Laboratory and the Iowa Agricultural and Home Economics Experiment Station. The 12,535 corn borers captured in the four traps operated in 1964 was a 40-percent increase over 1963. The traps will be operated in 1965 with the same cooperators as in 1964.

#### M. Radiofrequency Treatment of Grain and Forage Seed

Previous studies have shown that all developmental stages of all stored-grain insects studied can be killed by exposure for a few seconds to RF fields of sufficient intensity. Such treatment does not damage the wheat for germination or milling and baking purposes.

Last year equipment was constructed to pulse-modulate the output of the RF power oscillators in efforts to improve the efficiency of the method for insect control. Extensive studies were conducted this year to determine the most effective combinations of pulse repetition rate, pulse width, and field intensity. With pulse modulation, field intensity can be nearly twice that possible with continuous oscillation, but anticipated increases in insect mortality were not achieved generally. Pulse modulation did increase the mortality of lesser grain borers treated in wheat, however. Comparison of continuous and pulse-modulated treatments will be continued for different stored-grain insect species.

Studies were continued in cooperation with the Department of Entomology, University of Nebraska, in efforts to explain physiological effects of RF treatment on insects. Changes in weight loss and respiration following treatment were observed for yellow mealworm larvae which were exposed to sublethal treatments. Protein synthesis has been studied as a measurement of metabolic activity by incorporation of radioactive amino acids.

Electron microscopy studies on tissue from wax moth larvae revealed vacuolation in nerve cells in RF-treated insects which was not evident in untreated insects.

A pilot plant for continuous-process RF treatments was constructed this year. Initial results verified the exposure levels needed for stored-grain insect control which were previously determined in the laboratory with small samples.

Earlier studies have shown that RF treatments are effective in reducing the percentage of hard seed in alfalfa, red clover, ladino clover, and sweet-clover. Infrared and RF treatments were compared this year on more sweet-clover seed lots containing high hard-seed percentages. Both treatments reduced hard-seed content and provided a corresponding increase in germination. Both treatments were more effective when the seed moisture content was low. Twenty-seven alfalfa seed lots with high hard-seed content were collected from the Pacific Northwest and treated to learn how commercial seed lots will respond to RF treatment for hard-seed reduction. Treated samples are now being tested for germination.

Low germinating seed lots of Indian ricegrass and Cicer milkvetch were exposed to RF treatment, but the treatments employed were not effective in increasing germination.

At Pullman, Washington, sweetclover seed samples at nine moisture contents ranging from 2.0 percent to 15.4 percent were irradiated in the glow-discharge chamber. Results indicated that the seeds could withstand a higher level treatment with each successively lower moisture content. Any reduction in hard-seed content in samples with higher than 4 percent moisture was accompanied by an increase in the number of dead seeds. At lower moisture content, treatment levels were found which increased the germination percentage by reducing the hard-seed content. Studies on sweetclover seed will be continued.

A continuous-treatment glow-discharge chamber was used to irradiate alfalfa seed samples. Data obtained indicated that a significant reduction in hard-seed content could be accomplished in 30 seconds or less. The repeatability of results was not good. Temperature data were collected for various treatment conditions and attempts to correlate those with treatment results will be made.

Equations were written which related treatment temperatures to current, absolute pressure, and length of treatment for alfalfa seed in the batch-type glow-discharge chamber. Maximum treatment temperature was found to be a



reasonably good criterion for predicting significant reduction of hard-seed content in alfalfa. For seeds at 6 percent to 9 percent moisture a maximum temperature of 75° C. to 85° C. produced good results. Seeds at approximately 2 percent moisture were able to withstand about a 30° C. higher maximum temperature without a reduction in viability.

#### N. Electric Equipment for Soil Warming for Plant Growth

Investigations were continued in Indiana, and initiated in Maryland and Minnesota, to determine the requirements for installation and management of electric soil-heating cable systems to maintain suitable turf conditions for activities in critical-use areas during cold weather. In Lafayette, Indiana this work is cooperative with the Purdue University Agricultural Experiment Station through the Agronomy and Agricultural Engineering Departments.

Cold season soil warming has become eligible for acceptance as a part of turf management programs. Heat applied to the root zone of bluegrass plants altered the natural temperatures in which the turf existed, thereby keeping the soil from freezing, promoting root growth and blade extension, keeping turf greener, helping sod knit, and melting snow. The effectiveness of heat treatments was evaluated considering electric energy used, temperatures, turf condition and turf growth. Five heated plots and an unheated control area were studied. Each plot was 10 x 120 feet separated by 10 feet of unheated area. The study was conducted in the Purdue University varsity football practice field.

The warmest heated area did not freeze or crust during the 1963-64 winter. The control area was frozen 50 days during January, February and March of 1964. Two heated areas, each sustaining healthier areas than the warmest area, were frozen only 4 and 9 days during this interval. Control area temperatures averaged about 20° colder than the warmest treatment. Active growth, evidenced by blade and root extension, was observed during the entire winter months on the warmest heated area. Two of the intermediate level heated areas sustained healthy, vital, green turf during the winter months, but not active growth. In the remaining two treatments the turf grew later into fall and began growing earlier in the spring than the unheated areas.

Air temperature was found to be the best indicator of when heat should be applied. Temperatures in the soil defined the heat reserve and soil thermostats were used as limit switches to maintain the desired soil warmth. Use of time clocks allowed inclusion of off-peak electrical demand considerations.

Installations were made at Beltsville, Maryland and St. Paul, Minnesota, in order to study the effectiveness of soil heating in locations that have different winter weather conditions.

## O. Solar Grain Drying Equipment

Solar grain drying equipment has been developed in Kansas. Equipment using plastic collapsible solar collectors and fans used only 40 percent as much electric energy for fan operation as a system using fans without solar collectors.

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## GRAIN AND FORAGE CROPS RESEARCH

of the

United States Department of Agriculture  
and related work of the  
State Agricultural Experiment Stations

### Section B

This progress report of U.S.D.A. and cooperative research is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on U.S.D.A. and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of U.S.D.A. and cooperative research issued during the last year. Current agricultural research findings are also published in the monthly U.S.D.A. publications, Agricultural Research, Agricultural Marketing, and The Farm Index.

UNITED STATES DEPARTMENT OF AGRICULTURE  
Washington, D. C.  
December 15, 1965

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## II. NUTRITION, CONSUMER AND INDUSTRIAL USE RESEARCH

### CEREAL STARCHES - INDUSTRIAL UTILIZATION

Northern Utilization Research and Development Division, ARS

Problem. Starch accounts for about two-thirds the weight of all grains.

Finding new, large-volume outlets for starch would, therefore, result in substantially increased consumption of cereal grains. Of the 5.6 billion pounds of cereal starch now produced, about 3.5 billion pounds is used ultimately in food products, and increases would be expected to follow population growth. However, the remaining 2.1 billion pounds find industrial outlets that offer opportunities for increases at a rate greater than that of population growth. Because starch must compete with products derived from nonagricultural sources, these opportunities can best be realized by a program of research designed both to maintain the competitive position of starch in its current uses and to develop economical new industrial uses.

Substantial new outlets for cereal starches and flours can be envisioned if basic research and development on several types of chemical and physical modification of starch and flour now in the experimental stage or anticipated can be prosecuted to successful conclusion. This research is mainly directed toward new products for the pulp and paper industries and for the building and construction industries, but other industries, such as the chemical, petroleum, mining, textile, plastics, coatings, and packaging industries, also provide attractive opportunities. New outlets for starch that appear very promising include use of modified starches as wet-strength additives for paper, water-resistant adhesives, coatings, and foamed products, and of starch-derived pulps as an integral part of high-quality paper. In addition, if the competitive position of starch is successfully maintained through improvement by research, additional consumption would be expected by 1975 from participation in the multimillion-bushel markets for grain resulting from normal growth of existing industrial outlets for starch and flour such as paper, textiles, packaging adhesives, drilling muds, and building materials.

To accelerate realization of these goals, more information is needed on the physical and chemical properties and chemical reactions of cereal starches, on economical methods for effecting desired physical and chemical modifications and on product evaluation and development. In addition, still further new markets for cereal starches should be possible from an adequate program of fundamental and exploratory research to discover new concepts, principles, and reactions leading to new processes and products for future development.

### USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing, long-range program of research involving analytical, organic and physical chemists and chemical engineers engaged



in basic, applied and developmental studies on the chemistry of cereal starches and their conversion to useful industrial products.

The Federal scientific effort for research on cereal starches totals 62.3 professional man-years. Of this number 19.1 are devoted to chemical composition and physical properties and 43.2 to new starch chemical derivatives and their evaluation.

Research at Peoria, Illinois, on chemical composition and physical properties (13.5 professional man-years) involves fundamental research on reactions of maltose and glucose, on amino acid and peptide derivatives of carbohydrates, and principles of graft polymerization. Research contracts (2.1 professional man-years) are in effect with the Arizona Agricultural Experiment Station, University of Arizona, Tucson, Arizona, for basic studies on the reaction of starch with mercaptans (.6 professional man-year) and on the reaction of acetylene with methyl glucoside (.6 professional man-year); with The Johns Hopkins University, Baltimore, Maryland, for basic research on the reactions of starch in fluid dynamic media (.7 professional man-year); and with Southern Illinois University, Carbondale, Illinois, for investigations on synthesis of maltooligosaccharides (.2 professional man-year). Contract research on the reaction of starch with acetylene was completed at the Arizona AES, University of Arizona, Tucson, Arizona. Grants (3.5 professional man-years) have been made to Ohio State University Research Foundation, Columbus, Ohio, for basic research on the reaction of vinyl ethers with carbohydrates (1.4 professional man-years); to Ohio State University, Columbus, Ohio, for basic investigations of unsaturated and sulfur-containing carbohydrates (.8 professional man-year); and to Purdue Research Foundation, Lafayette, Indiana, for studies on sugars containing carbon-bound nitrogen, phosphorus and sulfur (1.3 professional man-years).

Research at Peoria, Illinois, on new starch chemical derivatives and their evaluation (28.6 professional man-years) involves basic and applied studies on various types of chemical products derived from starch and dextrin and in evaluation of these products for various industrial uses such as pulp and paper products, plastics, coatings, organic chemicals and stable viscosity agents. Research contracts (14.6 professional man-years) are in effect with Ohio State University, Columbus, Ohio, for research on synthesis of amino derivatives of starch (1.2 professional man-years); with Battelle Memorial Institute, Columbus, Ohio, for developmental research on starch and other cereal grain xanthides (6.2 professional man-years) and for studies on starch derivatives for use as colloids in water-emulsion paints (1.2 professional man-years); with Stanford Research Institute, Menlo Park, California, for research on graft copolymers of cereal starches with vinyl-type monomers (1.3 professional man-years) and on process development of selected graft copolymers (2.3 professional man-years); with Archer Daniels Midland Company, Minneapolis, Minnesota, for evaluation of starch polyol urethane foams (2.0 professional man-years); and with Western Michigan University, Kalamazoo, Michigan, for evaluation of modified cyanoethylated starches for applications in paper (.4 professional man-year). During the reporting period, research

at the University of Minnesota, St. Paul, Minnesota, on reactions of dialdehyde starch in solution was completed.

The Department also sponsors research on cereal starches conducted by foreign institutions under grants of PL 480 funds. Research on chemical composition and physical properties involves grants to National Institute of Agronomic Research, Paris, France, for research on changes induced in starch by gamma irradiation (4 years, 1961-1965); "Giuliana Ronzoni" Scientific Institute for Chemistry and Biochemistry, Milan, Italy, for research on glucopyranose rings in starches and dextrans (5 years, 1962-1967); Institute for Fibres and Forest Products, Jerusalem, Israel, for studies on the mechanism and products of mild oxidation of starch (5 years, 1963-1968); University of London, London, England, for research on debranching enzymes and their use in studying the fine structure of starch components (5 years, 1963-1968); University of Osaka Prefecture, Osaka, Japan, for development of an analytical method for carbonyl groups in carbohydrates (4 years, 1964-1968); and University of Edinburgh, Edinburgh, Scotland, for studies on the mechanism and structural changes involved in thermal, acid and alkaline degradation of starches (5 years, 1964-1969). During the reporting year research was completed on starch structure as revealed by interaction of starch and enzymes at the University of Birmingham, Birmingham, England, and on the proteolysis inhibiting effects of cereal starches and flours at the National Institute of Hygiene, Paris, France.

Research on new starch chemical derivatives and their evaluation involves grants to Hebrew University, Jerusalem, Israel, for studies on starch vinyl and epoxide graft copolymers (4 years, 1963-1967); Institute of Industrial Chemistry, Bologna, Italy, for studies on fatty chemical derivatives of starch dextrans (5 years, 1960-1965); National Institute of Technology, Rio de Janeiro, Brazil, for research on phosphorus- and sulfur-containing cationic starches (5 years, 1962-1967); Ahmedabad Textile Industry's Research Association, Ahmedabad, India, for research on starch-gum copolymers prepared by codextrinization (5 years, 1963-1968), and for studies on preparation and characterization of hydroxyethyl ethers of cereal starches (5 years, 1965-1970); Academy of Sciences and Chemical Institute "Boris Kidric," Ljubljana, Yugoslavia, for studies on modification of starch by moisture and temperature treatments (5 years, 1964-1969); and Plastics Research Institute TNO, Delft, The Netherlands, for research on preparation of metal alkoxides of starch for use as intermediates in synthesis (5 years, 1964-1969).

#### PROGRAM OF STATE EXPERIMENT STATIONS

State stations conduct a continuing program of research on the fundamental chemistry of cereal starches and their utilization. One regional project, NC-60, is directed to modification of starch for industrial uses. Participating States are seeking to: determine the fundamental reactions in the nonenzymatic dextrinization of starch; investigate chemical polymerization of D-glucose derivatives for the production of new types of synthetic



polymers; determine the mode of action of oxidants on starch; modify the basic structure of the D-glucose units in starch; discover enzymatic reactions which can modify starch and the effect of structural characteristics of starch on the action of enzymes; and develop methods by which nitrogen can be chemically attached to starch.

Other basic research is directed to study of the fundamental structure of complex carbohydrates and the mechanism of their formation and breakdown. Enzyme systems from plants and microorganisms are being examined from the viewpoint of their effect and role in structural changes, biosynthesis and deposition of starch. Systems of pure carbohydrases capable of attacking the whole spectrum of polysaccharide structure are being sought and their nature, mechanism and specific actions are being determined. Improved methods for separating and purifying a number of dextrin fractions continue to be studied. A new method for enzymatic synthesis of amyloextrins is being pursued.

Production of new types of hydrophilic polymers offers additional research challenges. Introduction of mercapto and amino groups as well as anhydro bridges in place of hydroxyl substituents is leading to additional families of modified polysaccharides. The resulting polymers are being characterized physically and biochemically.

Advances in carbohydrate chemistry continue to yield new and improved synthetic methods for preparing derivatives of both mono- and complex polysaccharides such as sulfated polysaccharides, heterocyclic sulfur sugars, new preparations of gentiobiose, more convenient introduction of isopropylidene groups, and novel syntheses of phosphorylated sugar acids. Studies of the reactivity and micro-structure of starch granules illustrates still another facet to the starch utilization program of the stations.

The total scientific effort of the State experiment stations is about 7.1 professional man-years devoted to this area.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Chemical Composition and Physical Properties

1. Reactions of maltose and glucose. Deacetylation of 1-aminoglucose pentaacetate and 1-aminomaltose octaacetate with methanolic ammonia under identical conditions gave the N-acetyl derivatives of 1-aminoglucose and 1-aminomaltose, respectively. The latter retained 1 mole of acetamide as a crystalline addition compound that could be decomposed by boiling absolute ethanol but not by boiling ethyl acetate. Urea and 2-oxazolidone were shown to form 1:1 addition compounds with D-glucose and maltose that can be used to direct the course of chemical reactions of these sugars to form new derivatives. Thus, acetylation and alcoholysis proceed differently with the complexes than with the free sugars. The complexing amide does not react to combine with the products. Further instrumental examination confirmed that



alcoholated hydroxide ion is the absorbing species involved in near-infrared determination of hydroxide ion in aqueous and nonaqueous alcohol. Other studies showed that the enhanced optical rotation of  $\beta$ -methyl glucoside in alcoholic sodium and other alkali-metal hydroxides is not due to alkoxide formation but more probably is caused by distortion in the cyclic conformation resulting from adduct and chelate formation.

A method for preparing mixtures of C-4 methylated maltooligosaccharides was worked out at Southern Illinois University. Also, a series of 4-O-methyl maltooligosaccharides of degree of polymerization 2 to 7 has been prepared.

2. Reactions of carbohydrates with acetylene and mercaptans. At the University of Arizona, research on vinylation of starch has been completed. In the final phase of the work attempts to polymerize vinyl starch with a cationic catalyst indicated crosslinkage through acetal groups rather than through polymerization of the vinyl ether groups. Films crosslinked after being cast were of poor quality. Initial studies at Arizona on the related vinylation of methyl glucoside were largely devoted to development of needed separation and analytical procedures and to synthesis of needed reference compounds. A new monovinyl compound has been isolated and characterized as methyl 3-O-vinyl- $\alpha$ -D-glucopyranoside.

In the research on polymeric mercaptals, also in progress at the University of Arizona, no reaction was observed between 1,10-decanedithiol and starch or, more unexpectedly, dialdehyde starch. Statistically designed experimentation provided useful information on the relationships among conditions, extent of reaction and properties of products in the reaction between glucose and dimercaptans.

3. Amino acid and peptide derivatives of starch. Treatment of methyl  $\alpha$ -D-glucoside with sulfur monochloride selectively produced the 6-chloro-6-deoxy derivative. Glucose and carbobenzoxy-glycine activated by N,N-dicyclohexylcarbodiimide yielded a resolvable mixture of two mono- and one di-(aminoacyl) glucose derivatives. Studies were conducted to obtain further basic information on the influence of structure in the use of  $S_2Cl_2$  to replace hydroxyl groups with chlorine. This research on the synthesis of chloroglucose derivatives is directed to the possibility of alkylating protein nitrogen with a chloro-sugar. Successful preparation of specific aminoacyl sugars will enable needed determination of their properties.

4. New derivatives of starch and related carbohydrates. Under the grant at Ohio State University for research on unsaturated and sulfur-containing derivatives, two methods have been devised for introducing a double bond in place of *cis*- or *trans*-diol groups in glucose derivatives. This research has been strengthened by a grant to Purdue Research Foundation for research on new derivatives containing nitrogen, sulfur or phosphorus. In other studies at Ohio State, preliminary results of research on the reaction of vinyl ethers with carbohydrates indicate that no single hydroxyl group of a sugar reacts preferentially.

5. Basic studies on graft polymerization. Research on graft copolymerization comprised studies on the effects of reaction conditions, on fractionation of reaction products and on development of a method for enzymatic hydrolysis of the starch portion of the graft copolymer. Replication of the ceric-ion catalyzed grafting of acrylonitrile to gelatinized wheat starch showed that reproducibility was good. Molecular weight of the grafted chains ranged from 710,000 to 840,000 and graft site frequency was 3,940 to 4,600 anhydroglucose units per graft chain.

Partial hydrolysis of these starch-acrylonitrile graft copolymers is the obvious route to obtain water-dispersible products for industrial applications. The reproducibility of the graft copolymerization reactions indicates that final products having acceptably constant properties from batch to batch should be obtained without great difficulty.

6. Application of nuclear magnetic resonance to starch structure and chemistry. This newly undertaken research is designed to utilize nuclear magnetic resonance, a powerful new instrumental technique for characterization of organic molecules, in the solution of problems relating to the chemistry and structure of starch, starch components and their chemical derivatives. These studies already have provided good evidence for definite complex formation between OH groups and certain proton acceptors. Observations were made with the aid of an instrument which minimizes transient signals or "noise." The results strongly indicated the existence of the helical configuration for amylose in solution. Previously the only evidence for this configuration was derived from X-ray patterns of solid materials or deduced from chemical reactivity. Unlike NMR, older conventional procedures could not be used effectively for measurements on dilute solutions.

7. Reactions of starch in fluid dynamic media. Encouraging results have been obtained in initial trials at The Johns Hopkins University of the fluidized bed reactor for converting starch to levoglucosan (see previous annual report). Variation of the dielectric constant of starch with frequency was found to be too small to serve as the basis of a practical analytical method for starch.

8. Proteolysis inhibition by starch. Under a PL 480 grant at the National Institute of Hygiene, Paris, France, studies have been made on the physiological activity of concentrates of wheat antitryptic factor. Protein digestion in rats was inhibited by high levels of the factor. Laboratory work on this project has been completed. The final report, which has not yet been received, is expected to provide information on the nature of this effect on digestion and its possible relationship to nutrition.

9. Starch structure and degradation. At the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, scientists working under a PL 480 grant have completed phases of research involving infrared spectra of amylose and its oligomers and hydroxyl proton resonances of sugars



in dimethyl-sulfoxide solution. This research is part of detailed investigations on the glucopyranose ring structure of starch.

In studies under a PL 480 grant at the University of Birmingham, Birmingham, England, theoretical equations were derived to explain sedimentation and electrophoresis patterns of interacting polymers, the equations being applied specifically to  $\beta$ -lactoglobulin. A method was developed for obtaining synthetic amylose having a high molecular weight, no branches, and a narrow size distribution. The product may be a useful model for amylose. These studies have been completed, but the final report has not yet been received.

At the University of London, London, England, under a PL 480 grant for research on starch- and glycogen-debranching enzymes, a method has been devised for determination of the average chain length or degree of branching of amylopectin by its complete enzymatic hydrolysis with  $\beta$ -amylase and pullulanase, followed by measurement of the amount of glucose in the resulting mixture of maltose and glucose. A branching enzyme has been isolated from sweet corn endosperm and its mechanism of action has been determined.

Studies on thermal degradation of starch and its components have been initiated at the University of Edinburgh, Edinburgh, Scotland. Results so far indicate the absence of an induction period or autocatalysis in the decompositions.

10. Mild oxidation of starch. In research under a PL 480 grant to the Institute for Fibres and Forest Products, Jerusalem, Israel, study of the oxidation of starch by bromine in acid medium has revealed the existence of a bromine-starch complex. Another interesting discovery is that the iodine affinity of wheat starch is increased by disintegration of native aggregates in acid and neutral solution.

## B. New Starch Chemical Derivatives and Their Evaluation

1. Chemical studies on starch xanthates and xanthides. Laboratory experiments showed that starch xanthate (D.S. 0.3) analyzed 10 minutes after discharge from the Ko-Kneader had 80 percent of the xanthate groups on secondary hydroxyls and 20 percent on primary hydroxyls. On aging for 3 days the xanthate groups approached equal distribution on the primary and secondary positions. When a highly viscous Ko-Kneader xanthate (from reactant sufficient to give D.S. 0.5 and 50 percent solids) was aged in sealed containers, the D.S. increased from an initial value of 0.29 to 0.43 in 4 hours and then decreased to near 0.29 in 6 days. Initial xanthate distribution favored the secondary position, but after a few hours 75 percent of the xanthate groups occupied primary positions.

Oxidation with lead tetraacetate was shown to be a useful tool for study of xanthides and other sulfur derivatives or carbohydrates. The structures  $(RO)_2C=S$  and  $(RS)_2C=S$  are oxidized by one mole of the tetraacetate to the



structures  $(RO)_2C=O$  and  $(RS)_2C=O$ , respectively, whereas the  $-SH$  group is converted by one-half mole to  $-S-S-$ .

A series of experiments on the effect of reaction conditions on the oxidative coupling of starch xanthate using chlorine indicated that, except for minor compensable differences, chlorine and hypochlorite display equivalent effectiveness.

2. Process and product development research on starch xanthates and xanthides. Studies in cooperation with Forest Products Laboratory continue to indicate good potential for xanthide as a means of improving resistance of linerboard to high humidity. For corrugated board, starch xanthides (D.S. 0.07-0.25) produced significant improvement in flat crush and wet and dry tensile strength. On the other hand, in insulation board, use of cereal xanthates of D.S. 0.25 or less gave little if any improvement in wet strength.

Engineering research resulted in a process for xanthation of starch based on the use of starch pastes of sufficient fluidity to permit transport and mixing with sodium hydroxide solution and carbon disulfide by means of ordinary pumps and other conventional equipment. Xanthates prepared by this method were found to be equivalent in handsheet tests to those of the same D.S. prepared in the Ko-Kneader. Further investigation revealed that trisodium phosphate (TSP) was sufficiently alkaline to replace sodium hydroxide. Use of TSP enhanced ease of precipitation of xanthides from solutions of xanthates having a D.S. as low as 0.03. Other studies provided data needed for design of a fully continuous xanthation process based on these findings.

In studies to find optimum methods for incorporating xanthate into paper, a continuous process for oxidative crosslinking of starch xanthate in pulp furnishes has been devised and is being scaled up to handle 5 pounds of pulp per minute. In preliminary tests, the process resulted in exceptionally high effectiveness of xanthide in improving the properties of paper.

The xanthation phase of the contract project at Battelle Memorial Institute on production of xanthides with larger scale equipment has been completed. At throughput rates of 100 to 200 pounds of starch and  $CS_2$ -starch mole ratios of 0.1 to 0.3, xanthation efficiencies for samples aged 5 minutes are similar to those observed at the Northern Division for samples aged 10 minutes to 1 hour after discharge from the Ko-Kneader. Removal of sulfur byproducts from xanthate dispersions was achieved by continuous countercurrent contact of air with the xanthate acidified to pH 6.

Battelle xanthates were found to require several times the amount of hypochlorite for in situ crosslinking that was established for xanthates prepared at the Northern Division. Production of satisfactory handsheets also presented problems. These difficulties have been resolved with the discovery that differences in pH and hardness of water were responsible for the discrepancies.

Colloidal ex situ crosslinked xanthides were prepared at Battelle by two methods. These products had activity as wet-strength agents paralleling that of in situ crosslinked xanthide. Although in their present state of development the methods used would not be commercially practical, the successful preparation of ex situ crosslinked xanthides comparable to in situ products in activity as wet-strength agents is a very important result in view of the probable greater industrial attractiveness of an ex situ crosslinked product.

The results of the extensive research program summarized in this and the preceding section show that excellent progress has been made toward development of economical and industrially attractive processes for preparing cereal xanthates and for incorporating them into paper products having optimum properties.

3. Starch polyol foams. Statistically designed experiments revealed the optimum conditions for the acid-catalyzed glycolysis of starch. A method was found for avoiding premature gel formation in this step. Replicate experiments showed that the subsequent reaction with propylene oxide formed polyethers with highly reproducible hydroxyl number ( $413 \pm 2$ ) and viscosity ( $44,000 \pm 1,500$  c.p.s.). Major components of the crude glycoside mixture obtained by reacting starch and ethylene glycol were shown to be glycol  $\alpha$ -D-glucoside (45-48 percent); glycol  $\beta$ -D-glucoside (21-24 percent), and glycol diglucosides (11 percent).

In the studies at Archer Daniels Midland Company on evaluation of plastic foams based on starch-derived glycol glucosides, laboratory-scale polyether preparations, foam screening tests and preliminary pilot-plant work were essentially completed. An improvement in the glucoside synthesis was devised that has the advantages of requiring less excess glycol and of avoiding gelation.

4. Graft copolymers. At Stanford Research Institute detailed studies showed that in grafting acrylic acid to pre-irradiated starch granules suspended in water, the reaction is complete in about 2 hours. A longer reaction time merely increased homopolymer formation. A study of solvents for graft copolymerization of starch and acrylamide showed that grafts with high acrylamide content were obtained with aqueous glycerol and sorbitol. The molecular weight of the grafted chains was higher with these two solvents than with the previously used aqueous ethylene glycol. High levels of add-on were obtained in the ceric-ion catalyzed graft copolymerization of starch with ethyl and butyl acrylates and methyl methacrylate, but not with methyl acrylate. A small pilot plant for continuous grafting of acrylonitrile to starch by ceric ion catalysts was designed and constructed. In the pilot-plant products, the grafts were shorter and more frequent than those of laboratory preparations. These changed characteristics in comparison to earlier laboratory preparations are highly desirable in terms of improved solubility, melt temperature and other important properties. A



100-pound sample produced in the continuous reactor was shipped to the Northern Division for evaluation.

In research under a PL 480 grant to the Hebrew University, Jerusalem, Israel, laboratory methods have been developed for the anionic graft polymerization of acrylonitrile, methacrylonitrile, methyl methacrylate, and propylene oxide onto starch and dextrin. The anionic method gives products with shorter and more frequent grafts than have been obtained thus far in other studies with free radical processes such as those described in the preceding paragraph. The anionically prepared products will therefore be most useful in relating graft size and frequency to properties. However, solvents other than dimethyl sulfoxide and tetrahydrofuran, those currently used in anionic grafting, will be required for a practical process.

5. Dialdehyde starch-protein plywood glues. The technique of preparing soyflour-blood-dialdehyde starch glue has been simplified and an improved formulation at lower cost has been achieved. In cooperative tests at Forest Products Laboratory, southern pine plywood bonded with this glue was essentially equivalent to pine plywood bonded with a commercial blood-phenolic resin glue. The experimental plywood has also shown good durability in 6 months of outdoor exposure. These results encourage the expectation that this new plywood glue will achieve commercial use.

6. New chemical products from starch and dextrin. The reaction of starch xanthate in water with amino compounds such as ethylene diamine, decamethylene diamine, and polyethylenimine yielded gels whose rigidity was found to depend on xanthate D.S., xanthate-amine ratio, amine molecular weight, and amount of water. Some of the gels displayed interesting adhesive and elastic properties that justify exploration of their potential as wet-end additives to paper as adhesives and sealants.

Starch xanthide and zinc starch xanthate have been compounded with natural and synthetic elastomers. The starch derivatives were compatible with the polymers even at the equal-weight level. Tensile strengths of the elastomers were increased several hundred percent by incorporation of 30-80 parts of cereal derivative per hundred parts of elastomer. Elongation and swelling in benzene decreased as starch content increased. There was little loss of starch derivative on extraction with hot water. In some experiments the starch derivative served as accelerator, curing agent and filler. Products varied from highly elastic materials to compositions resembling vinyl floor tile. Both of these discoveries--gel formation and compounding with elastomers--appear very promising and could lead to new outlets for cereal products.

A series of products was prepared by cyanoethylating cationic (aminoalkylated) starches. Cyanoethylation did not improve the properties of the cationic starches for various applications in paper. Detailed evaluation of low D.S. cyanoethylated starches for applications in paper manufacture are being undertaken in contract research by Western Michigan University. Contract



research to investigate starch derivatives for use in water emulsion paints will be performed by Battelle Memorial Institute.

In contract research on amino sugars at Ohio State University, derivatives of 3,6-diamino-3,6-dideoxy-D-altrose and of 2,6-diamino-2,6-dideoxy-D-mannose have been synthesized. Progress is being made in efforts to prepare 2-amino-2-deoxyamylose and 3-amino-3-deoxyamylose.

Under a PL 480 grant to the Institute of Industrial Chemistry, Bologna, Italy, significant progress was made in preparing, characterizing, and testing amino dextrans and fatty acyl dextrin derivatives. The amino dextrans, containing 0.1 to 0.5 percent nitrogen, are new products with adhesive properties which show potential uses as emulsifying agents in agricultural pesticide formulations and possibly also in printing inks and paper sizings. Also under a PL 480 grant, scientists at the Ahmedabad Textile Industry's Research Association, Ahmedabad, India, have carried out heat degradation of corn starch and gum karaya separately and in admixture in the temperature range 100-200° C. Evaluation at the Northern Division showed that a product from a mixture having a 9:1 ratio of starch to gum dextrin possessed some deflocculant ability.

Several cationic starch derivatives containing phosphorus and sulfur moieties have been prepared under a PL 480 grant at the National Institute of Technology, Rio de Janeiro, Brazil. Evaluation of the products at the Northern Division for cationic properties and for their fire-retarding properties in urethane foams gave disappointing results. Other research under PL 480 grants involves studies on preparation of metal alkoxides of starch at the Plastics Research Institute, TNO, Delft, The Netherlands; on hydroxyethylation of starch at the Ahmedabad Textile Industry's Research Association, Ahmedabad, India; and on modification of starch granules to obtain new paste properties at the Academy of Sciences and Chemical Institute "Boris Kidric," Ljubljana, Yugoslavia. No important results under these projects were reported during the past year.

7. Evaluation of starch derivatives in paper and paper products. Research on new chemical products from starch is supported by evaluation studies to determine the quality and performance of these products in applications in the pulp, paper and paperboard industry. During the reporting period tests were conducted, for example, with starch xanthates and xanthides and hetero-substituted starches. Results on the evaluation studies are reported in conjunction with the general discussion of research on the specific starch product. As a part of this work, variables involved in the crosslinking and handsheet procedures used in evaluation of starch xanthates and xanthides have been identified and can now be adequately controlled. The resulting improved reproducibility has resulted in more effective screening operations.

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## WHEAT - INDUSTRIAL UTILIZATION

Northern Utilization Research and Development Division, ARS

Problem. Wheat traditionally commands a higher price than corn. Since the starch content and starch properties of these two cereals are similar, new industrial uses for wheat must rely on advantages to be obtained from other components. Wheat flour is a mixture of starch, protein, gums, fiber, and fat. Because of the simultaneous presence of these basic ingredients, opportunities are promising for development of a wide variety of industrial products from wheat flour that would be expected to have properties and uses different from those of related products derived from refined starch. The problem is to find means for economical modification and reaction of these ingredients with each other and with other chemicals in order to realize the potential of the combinations.

Basic research now being conducted by the Department points to new potential industrial uses for cereal starches and flours that could consume significant quantities of grain by 1975. Among potential outlets for wheat flour are sizes for many special grades of paper, cereal pulps that would form an integral part of such papers, and plastic or foamed compositions for hard-board and insulating boards. The opportunity for successful realization of these possibilities is enhanced by recently developed fine-grinding and air-classification milling techniques that permit the composition of flour to be varied over wide ranges. These techniques are now satisfactory for soft wheats, but ways must be found to adapt them to hard wheats which constitute over 90 percent of the wheat remaining after current needs have been met.

Wheat flour could achieve its share of potential new markets more rapidly, and discovery of additional new uses under both public and private research programs would be facilitated, if more information were available on the basic physical properties and chemical reactions of flour and its components, on tempering and milling techniques, and on processing methods for economical conversion of flour to desired end products.

## USDA AND COOPERATIVE PROGRAMS

The Department conducts a continuing long-range program of research involving analytical, organic and physical chemists, chemical engineers and structural biologists engaged in basic studies of the chemical and physical properties of wheat, flour, flour fractions, and protein components and in applied research leading to new and improved wheat products for industrial use.

The Federal scientific effort for research on industrial utilization of wheat totals 44.8 professional man-years. Of this number 16.9 are devoted to chemical composition and physical properties, 12.7 to industrial chemical products, and 15.2 to processing technology.

Research at Peoria, Illinois, on chemical composition and physical properties (12.6 professional man-years) includes separation, characterization and chemical reactions of the component proteins of wheat gluten. Research contracts (2.4 professional man-years) are in effect at Purdue University, Lafayette, Indiana, for fundamental studies of the alkaline desulfurization of gluten (.8 professional man-year); and IIT Research Institute, Chicago, Illinois, for investigation of chemical modification of polypeptides obtained by hydrolysis of gluten (1.6 professional man-years). During the reporting period studies on methods for controlled hydrolysis of gluten, conducted at IIT Research Institute, Chicago, Illinois, were completed. Grants (1.9 professional man-years) have been made to Marquette University, Milwaukee, Wisconsin, for studies on synthesis and hydrolysis of amino acid glycosides (.5 professional man-year); to Kansas State University, Manhattan, Kansas, for investigations on separation of enzymes and proteins by disc electrophoresis (.8 professional man-year); and to Purdue Research Foundation, Lafayette, Indiana, for research on the effects of disulfide bond cleavage on the structure of wheat and corn endosperm proteins (.6 professional man-year).

Investigations on industrial chemical products conducted at Peoria, Illinois, (7.1 professional man-years) involve preparation and evaluation of new types of water-soluble and water-insoluble flour derivatives for industrial use. During the reporting period research specifically directed to wheat flour xanthates and xanthides was terminated, and engineering studies on acid-modified flour were completed. Further studies on wheat-derived xanthates and xanthides are being continued under contract research (see below) and under in-house projects broadly applicable to starch and flour xanthates and xanthides (see Area No. 1). Research contracts (5.6 professional man-years) are in effect with Iowa State University, Ames, Iowa, for engineering studies on use of pneumatic fluidization to effect acid modification of flour (.9 professional man-year); with Battelle Memorial Institute, Columbus, Ohio, for studies on preparation of xanthates from wheat bran and ground whole wheat and their use in making bag and box paper (3.0 professional man-years); and with Brown Paper Company, Berlin, New Hampshire, for evaluation of acid-modified flour as a surface-sizing agent for paper (1.7 professional man-years).

Processing technology research at Peoria, Illinois, (17.6 professional man-years) involves studies on conditioning and milling of wheat, air classification of flours, and reduction of viable microorganisms and radioactive contamination in wheat flour. A research contract (1.2 professional man-years) is in effect with the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for investigations on varietal variations in kernel properties and milling and fractionation characteristics of wheat. A grant has been made to Iowa State University, Ames, Iowa, for basic research on heat, mass and momentum transport (1.4 professional man-years).



The Department also sponsors research in this area conducted by foreign institutions under grants of PL 480 funds. Research on processing technology involves a grant to the Cereals Research Station, Research Association of British Flour-Millers, St. Albans, England, for investigations on quantitative measurement of properties of wheat that change significantly during conditioning (5 years, 1961-1966).

#### PROGRAM OF STATE EXPERIMENT STATIONS

Station research on use of wheat for purposes other than food has been limited. Some work is being devoted to economic feasibility studies related to use of wheat as a livestock feed when prices are competitive with prices of feed grains. Consideration is also being given to the supply and flow of wheats of different quality.

The total professional man-years devoted to industrial utilization of wheat is .6.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Chemical Composition and Physical Properties

1. Characterization of wheat gluten proteins. Optical rotatory studies showed conclusively that gluten, glutenin and gliadin all contain  $\alpha$ -helix. Glutenin and gliadin contained 17 and 25 percent  $\alpha$ -helix, respectively. Gluten had 20 percent, which is near the expected average of the two fractions. Molecular weights of gluten and gliadin were determined with a membrane osmometer. Values for gluten varied from 50,000 to 220,000 and values for gliadin from 75,000 to 81,000, depending on conditions (i.e., use of urea or aluminum lactate buffer, varying ionic strength and pH). Lower molecular weights were obtained in the presence of urea than in its absence. In the presence of urea, high pH decreased the molecular weight of gluten but change in ionic strength did not. Changes in pH and ionic strength did not affect the molecular weight of gliadin in urea solutions.

Properties of reduced glutenin reoxidized at 5 percent concentration indicated that it had more intermolecular disulfide bonds and a more highly branched structure than native glutenin. The number of ionizable groups and their pK's were determined for whole gluten proteins. Indications were found that the conformation of these proteins depends on pH and is different in alkaline than in acid solution.

This research is being expanded through a grant to Kansas State University for research on disc electrophoresis as a method for large-scale separation of proteins and enzymes.

2. Chemical reactions of wheat gluten. Studies on reactivity of functional groups important to wheat gluten chemistry showed that in comparable polar and steric environments SH groups reacted with acrylonitrile 300 times faster

than did  $\text{NH}_2$  groups. The nature of the vinyl compound also importantly affected reaction rate. Thus, methyl vinyl ketone reacted with glycine 700 times faster than did acrylamide. Whole gluten was completely derivatized by reaction with butyl acrylate. Only the lysine residues in the protein were involved in this reaction. Under optimum conditions for balance between hydrazide formation and peptide cleavage in hydrazinolysis of gluten, 26 percent of the primary amide groups were replaced by hydrazide residues and 10 percent of the peptide bonds were cleaved. These results show that progress is being made toward the goal--preparation of proteins modified in specific and pre-determined ways to adapt them to particular industrial end-uses.

Final results of research on gluten hydrolysis at IIT Research Institute indicated uniformity of charge distribution for the hydrolyzate. This observation, together with the previous demonstration of uniform particle size, indicates that polypeptides in the hydrolyzates have definite promise as raw materials for possible industrial products. Research to further characterize and to investigate chemical modification of these polypeptides will be conducted under a new contract.

Studies at Purdue University confirmed the greater rate of sulfur liberation from commercial gluten as compared to laboratory-prepared gluten. Kinetic studies were initiated on the release of sulfide sulfur, hydrolysis of amide groups and hydrolysis of peptide linkages. Significant differences were observed in the rate and extent of sulfide release between gliadin and glutenin when treated at different temperatures with different concentrations of alkali. This research is being strengthened by a grant to Purdue University providing for basic chemical studies on disulfide bond cleavage, a reaction of fundamental importance to the studies on desulfurization.

A grant has been made to Marquette University for studies on synthesis and degradation of O-glycosides of hydroxy amino acids that form protein to carbohydrate linkages in glycoproteins.

## B. Industrial Chemical Products

1. Acid-modified flour. In the final phase of engineering research on acid-modified flour a pneumatic reactor was designed, operated and developed to a point permitting the flour to be heated to  $140^\circ\text{F}$ . with negligible loss of moisture. These studies are now complete. A detailed evaluation of acid-modified flour as a paper size will be carried out under a contract with Brown Paper Company.

2. Fluidization of flour. In contract research at Iowa State University agglomerated flour was observed to behave ideally in a fluidized bed. For ordinary flour, addition of small amounts of silica or calcium silicate was required to secure good fluidization. In other tests, the rate of adsorption of hydrogen chloride on flour was 3 g. per 100 g. of flour per minute under ordinary moisture and temperature conditions. Rates as high as 10 g. per



100 g. per minute have been noted under some conditions. The limiting factor appeared to be the amount of water already adsorbed by the flour. Hydrogen chloride was more strongly bound by flour than is water, and there was significant heat of adsorption. In addition to this work, further information was obtained pertinent to the final design of a cylindrical column device for fluidizing flour.

3. Chemical modification of flour. Studies were made of reagents such as alkylenimines and dialkyl aminoalkyl halides for dry-state preparation of cationic flours. Ethylenimine appeared very promising since flours treated at 98° C. for 4 hours with as little as 1 percent of this reagent gave the same test results as sizing agents as did several commercial cationic starches.

4. Flour xanthates and xanthides. During the reporting year research on xanthation and on the use of crosslinked xanthides in paper was directed primarily to studies of the products derived from starch. The results of these studies are reported under Subheading B-1 of Area 1.

Contract research on xanthates and xanthides from wheat and derived materials is being conducted by Battelle Memorial Institute. Studies have been directed to preparation and characterization of wheat bran xanthates and their evaluation as components of paper. Optimum reactant ratios for preparation of bran xanthate have been tentatively established for a process using a conical mixer rather than the Ko-Kneader type of mixer. Initial studies on stability of bran xanthates, their oxidative crosslinking and incorporation into paper, and properties of the resulting products have given encouraging results. Light and electron microscopy are being used to aid in the definition of bonding mechanisms in bran- and starch-xanthide papers.

5. Evaluation of wheat flour products for applications in the pulp and paper industry. Research on chemically modified wheat flour and related products is supported by evaluation studies to determine the quality and performance of these products in applications in the pulp, paper and paperboard industry. During the reporting period tests were conducted, for example, with enzymatically modified flour and whole ground wheat and with cationic, chemically modified flours. Results of the evaluation studies are reported in conjunction with the general discussion of research on the specific wheat flour product.

## C. Processing Technology

1. Fine grinding and air classification of wheat flours. When milled and fractionated under identical conditions, 31 wheat varieties displayed an inverse relationship between protein shifting and kernel hardness as measured by the surface area of flour produced in a standardized grinding procedure. Protein shifts for the several classes of wheat averaged 6 percent for durum; 14 percent for HRS; 26 percent for HRW; 57 percent for SRW; and 62 percent for club. Impact milled flours from HRW wheat showed greater



protein shifts than conventionally milled flours, but increased ash and fat content in the flour and fractions resulted.

Engineering studies were conducted to estimate the grinding costs for conversion of mill feeds to various products. The estimated cost per ton for maximum coarseness ranging from 75 to 250 mesh were: red dog, \$2.20-\$4.42; middlings, \$3.80-\$6.84; and bran, \$5.70-\$8.00.

Initial studies at the University of Nebraska on crosses of soft and hard wheats of different kernel hardness have shown considerable variation in milling, air classification, and protein quality characteristics.

2. Wheat conditioning. In estimation of endosperm breakdown in flours from tempered wheat, microscopic analysis was shown to be necessary only for the fraction having particles below 30 microns in diameter. Free protein and free starch were found only in this fraction. The fraction above 30 microns contained only endosperm fragments and its breakdown was satisfactorily estimated by means of a nonselective procedure such as the Micromerograph.

Comparative microscopic sizing of wheat starches and the corresponding flours revealed that wheat having starch with a higher proportion of relatively small granules yields a flour containing more free protein particles. This result indicates that protein-starch binding is less firm in small granules formed late in kernel development than in large granules formed earlier.

3. Reduction of viable microorganisms in flour and flour products. Studies on procedures for reducing microorganisms in flour showed that the simplest method for reducing counts to less than 500 per gram is merely to store flour at 115° F. for about 10 days. Flour properties were little affected by this procedure. Continuation of microbiological examination of flours and parent wheats from mills in various locations showed that the bacterial counts of flours from the 1964 Kansas-Nebraska crop averaged about one-third lower than for the 1963 crop. Fungal counts were higher for the 1964 crop than for the 1963 crop. Bacterial counts were considerably higher for the parent wheats than for the flours milled from them, whereas the reverse order was observed for fungi. Only low levels of coliforms, fecal streptococci and lactic acid bacteria were found in flours. These findings indicate adequate mill sanitation and suggest that flour is not the principal source of lactic acid bacteria responsible for spoilage of refrigerated dough products.

4. Reduction of radioactive contamination of wheat and milled products. In the research on reducing radioactivity of wheat, a one-step washing of the grain with warm 0.5 percent citric acid solution resulted in removal of 73 percent of the Sr-90--the best results to date. This procedure has also resulted for the first time in a reduction of the residual radioactivity of patent flour. Milling studies of many samples of different varieties of wheat from various locations showed that varieties of wheat grown under identical conditions at the same location differ considerably in initial Sr-90 levels. This difference is reflected after milling by different

radioactivity in the feed fractions, whereas the radioactivity of the flour fractions was about the same. These investigations involve cooperation of the Health and Safety Laboratory of the AEC and have as their objective the development of processing methods that would yield nonhazardous wheat products in the event of a nuclear emergency. Present levels of radioactivity in wheat are well within the safe limits established by public health authorities.

5. Quantitative measurement of wheat conditioning variables. In studies under a PL 480 grant to the Cereals Research Station, Research Association of British Flour-Millers, St. Albans, England, appreciable increases in protein shifting (i.e., increased endosperm breakdown) was observed for flours milled from whole grain extracted with polar solvents. A series of tempering and milling experiments indicated that plastic deformation of the protein component weakens protein structure and leads to breakage during subsequent regrinding. Since plasticity of the protein increases with moisture content, so also does breakdown of endosperm that has undergone plastic deformation.

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CORN, SORGHUM, AND OTHER FEED GRAINS -  
IMPROVED INDUSTRIAL, FEED AND FOOD PRODUCTS  
Northern Utilization Research and Development Division, ARS

Problem. Abundant quantities of corn, sorghum, and other feed grains are now available beyond those amounts required to satisfy current needs. Both domestic consumption and export potential of these grains could be increased by development of new products for use by industry or of improved foods and feeds. Industrially, increased use of corn and sorghum will be mainly dependent upon increased markets for starch. However, flours derived from these grains are mixtures of starch, protein, and minor amounts of other components. Such mixtures have promise as raw materials for conversion to adhesives, water-soluble coatings, plastic materials, and related products that should have properties and uses different from related products derived from refined starch or wheat flour and that should contribute independently to increasing industrial markets. Isolated protein components of corn and sorghum flours should be suitable raw materials for production of useful resins and films. To achieve these utilization goals, more information is needed on basic physical and chemical properties and reactions of these flours, on the properties of component lipids, waxes, and proteins and their possible interactions with starch, and on the use of fine grinding and air classification and other new milling techniques for obtaining milled products having the most advantageous properties as industrial raw materials.

Because of the growing emphasis on increasing meat production, there is need for processes to obtain improved feed products such as high-protein feeds, mill feeds, feed concentrates, and feeds with high oil content. Such improvement could be achieved through research to obtain better knowledge of the biologically and nutritionally important constituents of corn, sorghum, and oats, to evaluate present, and to develop improved, milling and processing methods, and to ascertain the effects of such methods on the nutritional qualities of the products. In addition, because of the world shortage of protein in human nutrition, this research could enhance the export value of these grains by providing the necessary basis for development of high-protein and other food products that would be acceptable in foreign markets.

USDA AND COOPERATIVE PROGRAMS

The Department has a continuing long-term program involving analytical and organic chemists, chemical engineers and structural biologists engaged in basic studies of the components of corn and sorghum and in application of the new knowledge gained to the development of improved processing technology leading to more effective utilization of these cereal grains.

The Federal scientific effort for research in this area totals 8.0 professional man-years. Of this number 5.6 are devoted to chemical composition and physical properties and 2.4 to processing technology.

Research on chemical composition and physical properties (3.5 professional man-years), conducted at Peoria, Illinois, involves investigations of physiologically active nonprotein nitrogen substances in corn and of carotenoid pigments of corn, corn milling fractions, and yellow endosperm sorghum. A portion of the effort on carotenoid pigments is cooperative with Crops Research Division and is directed to development of corn and sorghum varieties having high carotenoid content. Such varieties are needed for improved food and feed products and to enhance the competitive position of U. S. corn in international trade. Research contracts (2.1 professional man-years) are in effect with Indiana University Foundation, Bloomington, Indiana, for studies on the isolation and characterization of phenolic pigments of grain sorghum (.8 professional man-year); and with Kansas State University, Manhattan, Kansas, for investigations on the composition, processing and feeding value of hybrid grain sorghums (1.3 professional man-years).

Processing technology research conducted at Peoria, Illinois, (2.0 professional man-years) involves pilot-plant studies of conditions and methods for increasing the yield of oil and grits by dry-milling processes. Effects of processing variations on industrially and biologically important components of corn are determined. A grant has been made to Pennsylvania State University, University Park, Pennsylvania, for basic studies on the mechanical and viscoelastic properties of shelled corn as related to the corn dry-milling process (.4 professional man-year).

The Department also sponsors research in this area conducted under grants of PL 480 funds to the following foreign institutions: National Institute of Agronomic Research, Paris, France, for basic studies of the physical chemical properties of corn zein (4 years, 1961-1965); and Indian Institute of Science, Bangalore, India, for research on separation of grain sorghum proteins (5 years, 1963-1968). These lines of work are under the subheading chemical composition and physical properties. During the reporting period research was completed on oat antioxidants at the Cereals Research Station, Research Association of British Flour-Millers, St. Albans, England.

#### PROGRAM OF STATE EXPERIMENT STATIONS

State stations have a continuing program designed to improve the utilization of corn, sorghum, and other grains in feeds and foods. There is widespread interest in and much effort is devoted to development of varieties of grains with improved nutritive value, pigment content, or other special constituents of value in animal rations. Research on the chemical composition and physical properties of grains is being conducted to support the breeding program on the one hand, and the nutrition program on the other. Development and utilization of corn lines and hybrids with high-protein and high-oil content is an example of the work. The variation in fat and protein



content and in amino acid and fatty acid composition is being followed closely. Extraction and characterization of corn proteins permits selection of corn varieties with improved protein quality and provides information which may lead to new industrial uses.

The work on microbial problems associated with grain storage and utilization involves study of mold deterioration and its effect on wheat and corn. Biological changes are also investigated.

Handling, processing, storage and milling procedures are being researched both from the standpoint of improvement of processing procedures and from the standpoint of effects on ultimate utilization of the products. Processing treatments such as drying, pelleting, enzyme treatment, steaming, dry rolling, and pearling are being studied. Product form, such as whole, cracked, or pelleted, also affects maximum utilization of nutrients and feed efficiency. Product characteristics, such as moisture, protein and fiber content, and pigment retention are also evaluated in terms of value of the grains for feed.

The relative value of sorghum and corn for finishing beef steers is of great significance in the utilization of these grains. Effects of conditioning, drying procedures and storage conditions are important. Studies are in progress to determine the effects of processing grain-type sorghums on their utilization and nutritive value in beef and dairy rations.

Annual crop residues such as corn stalks, sorghum stems, and corn cobs provide a natural reservoir of raw materials. Studies on isolation, characterization and derivatization of hemicelluloses from these sources are in progress in an attempt to modify hemicelluloses in ways to produce new physical properties of potential usefulness.

Study of the economic feasibility and potential market expansion for selected grain crops through new uses and changed utilization patterns is in progress. Both agricultural and nonagricultural uses and particularly uses at processing and manufacturing levels are considered.

Total professional man-years involved in the utilization of cereals and other grains is 7.7

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Chemical Composition and Physical Properties

1. Nonprotein nitrogen substances of corn. Studies on nonprotein nitrogen components showed that most of the nucleotides of whole corn are in the germ. However, some endosperm nucleotides did not appear in the germ. Immature corn (milky stage) contained 10 times the concentration of nucleotides as did mature corn. Trichloroacetic acid was found to be a superior solvent for extraction of nucleotides from corn. Nucleotides definitely

characterized as components of whole corn are thymidine monophosphate, guanosine di- and triphosphates, uridine diphosphate, and adenosine triphosphate. The pyrimidine nucleotides are present in largest proportion (80 percent). Sugar derivatives of guanosine and cytosine diphosphates also were identified.

2. Carotenoids and other pigments of corn and sorghum. The principal carotenoid pigments of 11 genetically different corn samples were characterized and estimated quantitatively. Total carotenoids ranged from 0 to 57 p.p.m. As carotenoid content increased, zeinoxanthin decreased; cryptoxanthin remained relatively constant as a percentage of the whole; and lutein, zeaxanthin, and polyoxy compounds increased both in absolute quantity and as a percentage of the whole. At the 57 p.p.m. level, lutein and zeaxanthin represented 70 percent of the total carotenoid content. The maximum xanthophyll level in corn breeding samples furnished by Crops Research Division during the last 6 months has increased from 72 to 77 p.p.m., an increase of 7 percent. An increasing number of samples containing more than 60 p.p.m. of xanthophyll is being observed. The average for present commercial hybrids is about 25 p.p.m.

Research at Indiana University showed that sorghum pearlings (pericarp fraction) contained most of the pigments and other phenolic compounds. Hexane-extracted wax from the pearlings contained no phenols.

3. Corn and sorghum proteins. In studies under a PL 480 grant at the National Institute of Agronomic Research, Paris, France, analysis and characterization of zein proteins was continued. During the year, particular attention was given to measurement of molecular weight, rotatory dispersion, and sulfur-containing amino acids and to the application of gel electrophoresis, gel chromatography, and amino acid analysis to protein fractions.

Research on separation of grain sorghum proteins is in progress under a PL 480 grant to the Indian Institute of Science, Bangalore, India. Protein content of endosperm (dehulled seeds) and seed coat fractions has been determined for 44 genetic strains of grain sorghum from the Rockefeller Foundation World Sorghum Collection at New Delhi. The endosperm protein content ranged from 9.2 to 19.0 percent. The endosperm of four Indian varieties which were analyzed contained from 8.5 to 10.6 percent protein. Studies on extraction of the protein by several different solvents showed that highest recoveries were obtained with alcoholic alkali. Research on the detection and separation of component proteins has been initiated.

4. Antioxidants in oats. The identity of one group of phenolic antioxidants naturally present in oats has been established. These compounds have been identified as diesters of caffeic or ferulic acid with C-26 or C-28 long-chain fatty diols. A U. S. patent (assigned to the Department) has been granted on these antioxidants, which are comparable in activity to

propyl gallate. This research, now completed, was conducted under a PL 480 grant at the Cereals Research Station, Research Association of British Flour-Millers, St. Albans, England.

## B. Processing Technology

1. Corn dry milling. In studies on tempering low-moisture corn, it was found that temper times of 1 hour or more were needed for absorption of appreciable moisture by the germ and adjacent endosperm but not for good degermination. The principal value of tempering was in reduced production of fines and increased recovery of oil. A second temper mainly facilitated good hull removal. Preheating low-moisture (old or artificially dried) corn and temper water to 115° F. or higher before mixing or tempering with open steam practically eliminated stress-crack formation. Without prior warming, about half the kernels developed fissures, with resultant adverse effects on yield and oil content of large grits. A variety of samples has been provided for study under the grant at Pennsylvania State University on mechanical and viscoelastic properties of corn kernels.

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\*Research supported by PL 480 funds.



## HIGH-AMYLOSE CORN - INDUSTRIAL UTILIZATION

### Northern Utilization Research and Development Division, ARS

Problem. Varieties of corn have been achieved genetically that contain greatly increased amounts of amylose. Amylose, the linear fraction of starch, possesses film- and fiber-forming properties not available in ordinary starch which contains only about 27 percent of this component. Because the unique properties of amylose open areas of utilization closed to ordinary starch, the potential industrial value of this new crop is very high. Several problems must be solved, however, to realize this potential.

For high-amylose starch to have substantially improved properties as a raw material in comparison with ordinary starch, it should contain at least 80 percent of amylose. Many breeding samples have recently been observed that contain over 80 percent of amylose. However, only varieties containing up to about 75 percent have so far been commercially available. Over 5 million pounds of high-amylose starch from commercial plantings are now utilized annually by industry. Although breeding is the task of the geneticist, utilization research is needed to provide information on amylose content, on changes in quantities and properties of the amylose, amylopectin, and other components such as oil and protein, and on milling characteristics of breeding samples in order to insure availability of satisfactory varieties.

To insure utilization of the potentially large volume of high-amylose starch that could eventually become available, more information is needed on the chemical and physical properties of amylose and high-amylose starch and on methods for converting them economically to desired products. Success in this research could lead to an estimated consumption of over several hundred million pounds of high-amylose starch by 1975 in plastics, coatings, films, fibers, and related products to which the linear character of amylose could make contributions.

### USDA AND COOPERATIVE PROGRAMS

The Department conducts a long-term, continuing program of research involving analytical, organic and physical chemists, structural biologists, and chemical and mechanical engineers who are engaged in basic and applied research designed to increase knowledge of the properties and reactions of amylose and other components of high-amylose corn and to utilize this knowledge in development of attractive industrial applications for amylose and high-amylose starch.

The Federal scientific effort for research on utilization of high-amylose corn totals 13.1 professional man-years. Of this number 10.7 are devoted to chemical composition and physical properties and 2.4 to industrial utilization.

Research at Peoria, Illinois, on chemical composition and physical properties (9.1 professional man-years) involves study of amylose content of breeding samples, starch and starch granule composition, structure and properties; and composition and properties of proteins and other components of high-amylose corn. Studies on amylose content of breeding samples assist geneticists in developing varieties of high-amylose corn having increased amylose content. Cooperation with Field Crops Research Branch, Crops Research Division, is maintained in conducting these studies. A research contract (.2 professional man-year) is in effect at Arizona State University, Tempe, Arizona, for basic research on the interaction of "V" amylose with small organic molecules. Grants (1.4 professional man-years) have been made to the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for basic studies on variations in starch granules of genetically different corn samples (.8 professional man-year); and to Princeton University, Princeton, New Jersey, for basic research on the relationship of viscoelastic properties of amylose film to structure and function of plasticizers (.6 professional man-year).

Research on industrial utilization, which is conducted at Peoria, Illinois, is devoted to preliminary studies of chemical and physical modification of high-amylose starch as means for formation of amylose films having industrially acceptable properties (2.4 professional man-years).

#### PROGRAM OF STATE EXPERIMENT STATIONS

The station phase of the program designed to develop high-amylose corn for industrial uses is largely one of support for the breeding program. The Indiana, Missouri, and Nebraska stations are continuing development of lines high in amylose content. Samples are analyzed, often on an individual plant basis, to determine amylose content. Some additional effort is devoted to development of techniques or processes for separating amylose from other kernel constituents, to study of enzyme systems, and to search for industrial applications.

Total effort devoted to high-amylose corn utilization is about 1.0 professional man-year.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Chemical Composition and Physical Properties

1. Amylose content of breeding samples. During the reporting period 16,875 samples of high-amylose corn were analyzed. These were supplied under Memoranda of Understanding by the Bear Hybrid Corn Company, by the Missouri Agricultural Experiment Station and by Crops Research Division at the Missouri Station. Several thousand samples contained over 80 percent amylose. Among the samples analyzed during the last 6 months of the period were three samples at the 85-percent level, a new high in development of high-amylose corn. (In this report, percentages of amylose refer to



apparent values determined by iodine sorption. True amylose content, measured by quantitative fractionation, averages about 80 percent of the apparent value. According to reports from industry, this distinction has so far not proved significant in industrial utilization of high-amylose starch.)

In view of the attainment of strains having starch of 85 percent apparent amylose content, there is optimism that further progress will be made and belief that the limit of amylose content in high-amylose corn is not yet in sight. Limited quantities of Class 7A amylomaize (70-75 percent apparent amylose) have been grown by Bear Hybrid Corn Company and supplied to several processors for research purposes. It is anticipated that beginning with the 1967 planting season, Class 7 will be available to any processor requesting it.

2. Properties of components. The existence of protein-carbohydrate complexes involving amylopectin has been confirmed. In fact, it appears that such complexes exist in both the dispersed and granular state of amylopectin in dent and waxy corn. The complexes can be destroyed by phosphate buffer (pH 7), by hydrolysis at pH 4 and 100° C. or by action of the proteolytic enzyme Pronase. Based on the action of Pronase, a practical laboratory procedure has been developed that permits quantitative isolation of corn starch without degradation or alteration of the molecular structure of the starch components. Molecular weights of amylopectins from dent and 70-percent HA starches isolated by the new procedure are extremely high--1 billion and 175 million, respectively. The procedure is suitable for very small samples; e.g., starch content can be determined on as little as four kernels.

The availability of unaltered starch as provided by this new method of solution will greatly facilitate basic studies on starches from corn genotypes and should lead to much more accurate information on the properties and structure of these starches.

Contract research on V-amylose at Arizona State University was directed to investigation on the stability of V-amylose and V-amylose hydrate in equilibrium with salt-hydrates of known vapor pressure. At or above 50° C., the hydration reaction was reversible. The heat of reaction of water with amylose was calculated to be 9 k. cal. per mole of water. V-Amylose was reacted with gaseous and liquid NH<sub>3</sub> to form stable compounds. The reaction with liquid NH<sub>3</sub> is irreversible, suggesting that the NH<sub>3</sub> has penetrated to the interior of the helix and is tightly bound. The product retained the helical structure and was soluble in hot water.

Research on granule variation under the grant to the Nebraska AES indicates that water absorption of starch of the waxy genotype (wx) is much higher than that of starch from the sugary genotype (su2). Starch governed by the combination su2wx displayed an intermediate level of water absorption.



3. Molecular properties of amylose film. Molecular organization in amylose films cast from solutions in dimethyl sulfoxide (DMSO) was found to vary with the conditions of evaporation of the solvent. At 0-30 percent relative humidity amorphous structure resulted, as shown by X-ray diffraction. At 40-93 percent relative humidity, B and V structures were formed. Amorphous films could be converted to B-type by wetting with water and drying at room temperature. Heating amorphous films in a butanol-water mixture produced V-type structure. Agents used to plasticize amylose films, such as glycerol and ethylene glycol, formed recognizable complexes with the amylose.

Research on the mechanism of external plasticization of amylose films has been initiated under the grant to Princeton University.

#### B. Industrial Utilization

1. Chemical and physical modification of high-amylose starch. Treatment of HA starch with DMSO increased its reactivity to acid chlorides and anhydrides and other derivatizing reagents. Certain esters were soluble in hot water. Hydroxypropylated DMSO-pretreated HA starch (D.S. above 0.03) gave films soluble in hot or cold water. Unplasticized films had a tensile strength of 10,129 p.s.i., elongation of 11.4 percent and MIT fold of 523. Initial experiments showed that oxygen transmission of plasticized and unplasticized films was below the limit of measurement at 5° and 25° C. over a wide relative humidity range. Studies are planned to confirm these findings. The properties of these low D.S. products suggest that the cost of chemically modifying HA starch as a means of obtaining new or improved films or other products may turn out to be nominal.

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WHEAT AND CORN - FERMENTATIVE CONVERSION TO NEW  
INDUSTRIAL, FEED AND FOOD PRODUCTS  
Northern Utilization Research and Development Division, ARS

Problem. By fermentation of cereal grain substrates, new products can be obtained that are not readily available by other means and have promising potential for industrial, agricultural, and food uses. Processes now under development, if brought to successful conclusion, could lead to substantially increased consumption of grain for fermentative conversion to stable viscosity agents for secondary petroleum recovery by flooding of spent oil wells, to new organic acids and enzymes for industrial use, to feed supplements, and to effective biological insecticides and other pesticides that are harmless to man. In addition, there are good possibilities for utilizing fermentation processes to produce new food products that should promote foreign use of U. S. grains.

To accomplish these objectives and to realize the full potential of fermentative techniques for increasing utilization of grain, a broad program of exploratory research is required to find and identify through taxonomic studies species of organisms producing potentially valuable products, to isolate high-yielding strains or develop them by mutation, hybridization or genetic selection, and to develop basic information on culture media, special nutrients, and other factors required for optimum growth of microorganisms and maximum yields of desired products. Continued maintenance and expansion of a collection of pure cultures of well-characterized organisms is necessary for this research. For successful translation of laboratory results into commercially useful processes, more information is needed on new techniques of fermentation, on development of economical methods of growing organisms and handling fermentation processes on a large scale, and on special procedures for efficient isolation and purification of products from fermentative reaction mixtures. Finally, the most appropriate end uses for products must be identified and information obtained on product evaluation and development.

USDA AND COOPERATIVE PROGRAMS

The Department has a long-range continuing program involving analytical and organic chemists, biochemists, microbiologists, systematic biologists, and chemical engineers engaged in basic research on microorganisms and microbiological reactions and products and in application of both known and newly discovered principles to the development of practical fermentation processes for conversion of cereal grain substrates to useful chemical, biological, feed and food products.

The Federal scientific effort in this area of research totals 67.6 professional man-years. Of this number 29.4 are devoted to basic research on fermentation processes, 21.1 to industrial chemicals, 11.8 to biological pesticides, and 5.3 to feed and food products.



Basic research on fermentation processes conducted at Peoria, Illinois, (27.3 professional man-years) includes study of taxonomy of molds, yeasts and bacteria; factors affecting viability of microorganisms; and microbiological reactions and products. Because of the importance of the problem, the research effort on mycotoxins has been greatly intensified. Basic to these investigations and to the Division's entire research program on fermentation is assembly and maintenance in pure culture of a large collection of agriculturally and industrially important microorganisms. Much of the research on microbiological reactions and products is conducted by the Pioneering Laboratory for Microbiological Chemistry. During the reporting period taxonomic research on streptomycetes was restricted to studies required in connection with antifungal antibiotics. A research contract (.2 professional man-year) with American Type Culture Collection, Rockville, Maryland, provides for studies on preservation of certain microorganisms for which lyophilization is ineffective. A grant has been made to Iowa State University, Ames, Iowa, for investigation on bacterial amylases and their action patterns (1.9 professional man-years).

Research at Peoria, Illinois, on industrial chemicals (19.5 professional man-years) involves fermentative production of microbial gums, organic acids, and other products for use in the chemical industry. This work includes investigation and development of improved or new procedures for conducting industrial fermentations. During the reporting period studies on enzymatic modification of wheat flour were completed and replaced by research on enzymatic conversions of starch and glucose to products of possible industrial value. During the reporting period contract research on polymerization of selected fermentation acids and derivatives of fatty acids was completed at the University of Arizona, Tucson, Arizona. Grants (1.6 professional man-years) have been made to Cornell University, Ithaca, New York, for fundamental studies on biphasic fermentation (.8 professional man-year) and to the Nebraska Agricultural Experiment Station, University of Nebraska, Lincoln, Nebraska, for investigations on the nature of amylase enzymes (.8 professional man-year).

Research at Peoria, Illinois, on biological pesticides (9.1 professional man-years) is devoted to studies on biological insecticides for Japanese beetle and on antifungal antibiotics for use in control of economically important plant diseases. Investigations on biological insecticides for Japanese beetle and on other insect control agents is cooperative with Entomology Research Division and Plant Pest Control Division. Research on plant antibiotics involves cooperation with Crops Research Division. During the reporting period screening studies to find new antifungal antibiotics were completed and redirected toward practical evaluation of several new antibiotics revealed by the screening program. Research contracts (2.7 professional man-years) covering various phases of research on Japanese beetle pathogens are in effect at Michigan State University, East Lansing, Michigan, for basic research on enzyme activity in sporulation (.7 professional man-year); at Kansas State University, Manhattan, Kansas, for investigation of stabilization of vegetative cells of the pathogenic

organisms (.5 professional man-year); at the University of Minnesota, St. Paul, Minnesota, for fundamental studies on the transfer of genetic determinants of sporulation from one microorganism to another (.5 professional man-year); at the University of Illinois, Urbana, Illinois, for research on the applicability of a sporulation factor produced by bacteria to Japanese beetle pathogens (.6 professional man-year); and at Baylor University, Houston, Texas, for investigation of morphological changes involved in sporulation (.4 professional man-year). During the reporting period contract research providing for study of factors important to large-scale production of Japanese beetle pathogens was completed at Michigan State University, East Lansing, Michigan.

Research at Peoria, Illinois, on feed and food products (2.8 professional man-years) involves study of production of microbial carotenoids suitable for feed supplements and development of new fermented wheat foods that can help increase export markets for U. S. wheat. Research contracts (2.5 professional man-years) are in effect with A. D. Little, Inc., Cambridge, Massachusetts, for studies on stabilization of fermentative  $\beta$ -carotene (1.7 professional man-years); and with Consolidated Laboratories, Inc., Chicago Heights, Illinois, for research on the use of antimetabolites to facilitate selection of higher yielding strains of microorganisms producing  $\beta$ -carotene (.8 professional man-year).

The Department also sponsors research in the fermentation area conducted by foreign institutions under grants of PL 480 funds. Basic research on fermentation processes involves grants to the University of Milan, Milan, Italy, for basic studies on the metabolic pathway to 5-ketogluconic acid in Acetobacter species (5 years, 1961-1966); University of Allahabad, Allahabad, India, for collection of new Mucorales species (5 years, 1961-1966), and studies on survival of lyophilized microorganisms (5 years, 1962-1967); University of Durham, Newcastle-upon-Tyne, England, for investigations of sugar phosphate derivatives in molds (5 years, 1962-1967); Central Drug Research Institute, Lucknow, India, for studies on aerobic actinomycetes in India to find new accessions for the ARS Culture Collection (5 years, 1965-1970); and to the University of Liege, Liege, Belgium, for research to find lytic enzymes of microbial origin (5 years, 1964-1969). During the reporting period the project on Pseudomonas conversion of glucose at the Indian Institute of Science, Bangalore, India, was cancelled because adequate staffing could not be achieved. Research was completed on collection of new species of yeast at the National Institute for Agronomic Research, Madrid, Spain, and on organic phosphorus compounds of yeast at the University of Helsinki, Helsinki, Finland.

Research on industrial chemicals involves grants to the University of Lodz, Lodz, Poland, for research on the fermentative production of itatartaric acid (5 years, 1963-1968); University of Tokyo, Tokyo, Japan, for research on the fermentative production of D-tartaric acid (5 years, 1964-1969) and



of mevalonic acid (3 years, 1965-1968); and University of Baroda, Baroda, India, for studies on production of microbial lipases useful for modifying vegetable oils (5 years, 1965-1970).

Research on feed and food products involves a grant to the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, for research on production of vitamin B<sub>12</sub> (5 years, 1960-1965); and the National Institute for Agronomic Research, Paris, France, for studies on mutation of yeasts for improved feeds (3 years, 1963-1966).

#### PROGRAM OF STATE EXPERIMENT STATIONS

The Montana station is studying conversion of barley into feed yeast protein. Barley carbohydrates are converted enzymatically into fermentable sugars which, in turn, serve as an energy source for the yeast. Current work involves establishing reliable yield and cost data for analysis of the process. In fermentation studies at other stations, work is directed toward biosyntheses of lysine and of polysaccharides, for example. Still other research involves fundamental studies of the organisms, the fermentative process and methods for separating desired products from fermentation liquors.

The total research effort devoted to fermentative conversion as a means of utilization is 2.2 professional man-years.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Basic Research on Fermentation Processes

1. ARS Culture Collection. As of January 1, 1965, the ARS Culture Collection contained 16,934 permanent cultures, an increase of 2,863 over 1964. By way of comparison, the number of permanent cultures on January 1, 1960, was 8,883. The permanent collection has, therefore, nearly doubled in 5 years. During 1964 a total of 2,254 cultures were distributed to domestic and foreign recipients.

Contract research by the American Type Culture Collection revealed that 47 of 54 cultures (about 50 species) of molds tested remained viable after freezing and storing under liquid nitrogen for 2 weeks. This recently initiated work is designed to determine if the liquid nitrogen procedure can preserve organisms that do not survive lyophilization and have required serial transfer methods of maintenance.

Research of importance to the ARS Culture Collection is in progress at several foreign institutions under PL 480 grants. At the National Institute of Agronomic Research, Madrid, Spain, studies on collection and characterizing new strains of yeasts have been completed. More than 100,000 specimens were examined, and 1,399 cultures were characterized. A total of 1,023 yeasts were received and incorporated into the ARS Culture Collection. At



the University of Allahabad, Allahabad, India, results indicated that lyophilization of spores of Aspergillus niger did not adversely affect the production of citric acid. Also, there appeared to be a small decrease in viable spores as the lyophil preparations aged. Other research at the University of Allahabad is continuing to supply the Northern Division with many new isolates of Mucorales. A number of these cultures are being used in the in-house taxonomic studies on Rhizopus (see item 2, following).

2. Taxonomic investigations. A family of yeasts, the Chlamydozymaceae, has been erected based upon discovery of a highly developed and complex type of sexuality in an abundantly common group of yeasts whose asexual forms have been known for many years. The gametes consist of two sexes and the sexual reactions are strong, in some cases being driven by sexual agglutination. The distinguishing characteristic of protosexuality is the rapid conversion of bisexuals to unisexuals without an intervening stage that consists of sexual spores. These observations suggested the prediction that genera of protosexual yeasts would be found which had given rise to primitive ascomycetes and basidiomycetes at the taxonomic interface between the protosexual yeasts and two main classes of fungi that produce respectively ascospores and basidiospores. A very interesting species that occurs near this taxonomic interface was studied. The most primitive heterothallic species of Hansenula yet discovered, Hansenula wabatongushiensis, was found to have two sexual cycles, one ascosporeogenous, the other protosexual. The two phases are separately possessed by certain genetically different isolates.

In studies on Mucorales, new zygosporic strains have been found in two more heterothallic species in the section Dubiorugorhizopus of the genus Rhizopus. A (-) strain of R. niveus was found to give an imperfect mating with a (+) strain of R. oligosporus (the tempeh-producing organism). There are also one (-) strain of R. oligosporus and 12 (+) strains that can be successfully mated to give many mature zygosporic strains. These studies open possibilities for investigation of the effects of mating strains, sexual strains and possible hybridization on tempeh production and flavor.

The scope of basic taxonomic studies of streptomycetes was reduced soon after the start of the reporting year. In further investigations along these lines, emphasis is being placed on the taxonomy of special groups of antibiotic-producing organisms (see Section C, following).

3. Microbiological processes and products. In studies on genetic control of fermentation, omission of glucose from the medium or growth at 40° C. decreased but did not eliminate undesired autolysis of Pseudomonas aeruginosa. Several clones derived from one strain by combination of growth at 40° C. and exposure to acriflavin appeared to be free of autolysis. Attempts to secure conjugation between a male parent (strain 1093) and the female parent (1090) resulted in a preferential kill of the male parent. If mating was conducted at 40° instead of 28°, this kill became negligible.

A standard procedure was devised for evaluating the capacity of microorganisms to modify fatty acids fermentatively. Preliminary results indicated that degradation occurred with increasing difficulty as the chain length of the fatty acids increased. With availability of an effective method for evaluating the ability of microorganisms to modify fatty acids, accelerated progress is expected in identifying conversions of potential value to utilization research.

In the Pioneering Laboratory for Microbiological Chemistry, phosphatidyl glycerol and phosphatidyl ethanolamine were tentatively identified as two of three phospholipid fractions isolated from particles derived from bacterial cytoplasmic membranes and probably associated with electron transfer reactions in cell membranes and organelles of high metabolic activity. Studies on the macromolecules responsible for agglutination of certain yeast strains resulted in preparations with very high specific activity (agglutination achievable with 1.7 particles per yeast cell; 1.0 particle is the theoretical minimum). Specific activity increased with molecular weight over the range of 1 million to several hundred million. An acid phosphatase from Saccharomyces mellis was shown to be an exocellular enzyme. Research on the chemistry of compounds of microbial origin has shown that the acetylated hydroxy acid produced by an unidentified yeast is 8,9,13-triacetoxycosanoic acid.

In other work in the Pioneering Laboratory, the fungus Sclerotinia sclerotiorum was utilized as a model system for study of composition, metabolism and control mechanism involved in morphological development of sclerotia (hardened masses of mycelia resistant to unfavorable environmental conditions). Analysis of sclerotia of S. sclerotiorum showed that they are low in fat but contain large quantities of carbohydrate and protein. Several of the carbohydrates were identified. When sclerotia are placed in a suitable medium, they may either resume vegetative growth or develop sexual reproductive structures. Information was obtained on procedures for inducing the reproductive structures, on carbohydrate utilization by strains of S. sclerotiorum, on an extracellular polysaccharide produced in laboratory cultures, and on the biosynthetic pathway to D-mannitol in growing mycelium.

In studies under a PL 480 grant at the University of Milan, Milan, Italy, enzymes responsible for formation of 2-ketogluconate were found to be partly constitutive, whereas those producing 5-ketogluconate were inducible. Fermentations yielding primarily 2-ketogluconate may be possible if the basis for repression of the 5-ketogluconate enzyme system can be determined and controlled. Fermentations to yield only 5-ketogluconate appear to be impossible except by mutational loss of the constitutive 2-ketogluconate system.

At the University of Helsinki, Helsinki, Finland, all of the major metabolically active phosphate esters of yeast that accumulate or that are rapidly labelled with radioactive phosphate have now been identified.



Accumulation of trehalose phosphate in both Candida utilis and Saccharomyces cerevisiae was temperature dependent and enhanced by aeration. These observations suggest the presence of a thermally induced enzymic block. This research is being conducted under a PL 480 grant. Laboratory work has been completed, but the final report has not yet been received.

Studies recently undertaken under a PL 480 grant to the University of Liege, Liege, Belgium, have revealed conditions for obtaining high, reproducible, bacteriolytic activities in culture filtrates of Streptomyces albus G. A procedure for isolation and fractionation of the "F<sub>1</sub>" enzyme complex, which attacks several bacterial species resistant to lysozyme, has been developed. The F<sub>1</sub> muramidase has been separated from at least three less basic peptidases which have different pH optima and are species specific with regard to bacterial substrate.

Studies on microbial sugars, conducted under a PL 480 grant at the University of Durham, Newcastle-upon-Tyne, England, indicate that the ribitol teichoic acid from Streptomyces griseus is composed of ribitol residues joined through phosphodiester linkages.  $\beta$ -Glucopyranosyl substituents are attached to some of the ribitol units. The nucleotides in S. niveus have been identified as uridine diphosphate (UDP), UDP-glucose and a mixture of UDP-N-acetylglucosamine and UDP-N-acetylgalactosamine with a labile group attached to the amino sugar.

4. Aflatoxin investigations. Screening studies have revealed three strains of Aspergillus flavus to be the best aflatoxin producers among 22 strains tested. Aflatoxin could be produced on either still or shaken broths. Cracked corn, rice, wheat, sorghum and soybean grits were tested as substrates for aflatoxin production. Rice proved to be the best solid medium for producing consistently high yields of aflatoxin. The poorest medium was soybean grits with or without added methionine.

In the survey of commercial grains for incidence of aflatoxin, a total of 1,382 samples have been received and 347 samples from the poorest grades have been screened for aflatoxin. Positive results were obtained for 5 samples of sorghum, 3 of corn, and 11 of oats. Negative results were obtained for wheat. A number of soybean samples contained significant amounts of a fluorescing factor that proved not to be aflatoxin. These results are perhaps not unexpected since the low grades of grain tested are those to which moldy and damaged grains are assigned. Any conclusions regarding the incidence of aflatoxin in grains must await completion of the survey and study of data for the highest grades of grain. Growth of some species of the genus Bacillus and several Clostridia was inhibited when 10-20  $\mu$ g of 33 percent pure aflatoxin per ml. of substrate was present.

## B. Industrial Chemicals

1. Conversion of grains to fermentation media. A second strain of A. niger that produces high yields of glucamylase has been discovered. Formation of



transglucosylase is lower for this new strain than for the strain reported last year. Experiments with this earlier strain showed that amyloglucosidase yields of 10 to 12 units/ml. could be obtained in 20-liter fermentors with 20 percent corn slurry as substrate.

The organism Candida tropicalis Y-1367 was found to produce a transglucosylase capable of effecting reaction between starch or dextrin and glycerol. This enzyme could prove useful for large-scale production of glycerol glucoside.

2. Enzymatic modification of wheat flour. Tests of enzymatically modified ground whole wheat products showed that acceptable paper sizes could be obtained. Apparently because the action of the enzymes was affected by the presence of the bran, conditions for modification were markedly different from those required for flour. Settling of bran also constituted a problem. These investigations have been completed. Future effort will be directed to research on enzymatic conversions of starch and glucose to products having possible industrial value.

3. Studies on fermentation techniques. A two-stage stirred reactor comprising a 30-gal. fermentor in series with one of 60-gal. capacity was successfully used for a 6-day continuous fermentation to produce polysaccharide B-1459 in an overall yield of 42 percent. The product was of good quality, but foaming and viscosity characteristics indicated the possibility of physiological change in the culture. The polymer synthesized in the present studies has a lower uronic acid content and may be different in either molecular size, unit distribution or branching from batch-produced material.

Initial research on biphasic fermentation at Cornell University involved studies on the toxicity of water-immiscible alcohols to bakers yeast. In general, toxicity was proportional to solubility. Attempts were made to grow the B-1459 organism on membranes, but the growth and polysaccharide formation were not sufficiently better than in shake flasks to justify further study of this system.

4. Screening and structure studies on microbial polysaccharides. Gas chromatographic procedures have been developed for quantitative ( $\pm 1.0$  percent) resolution of mixtures of anhydroglucoses and of hexoses and pentoses. Phosphogalactans from Sporobolomyces sp. have been shown to contain galactose, phosphate and O-acetyl in the approximate molar ratio of 9:1:1. The phosphogalactosyl groups are terminal and permit galactose to be split off easily, leaving the monophosphate which still retains viscosity.

Studies on ultracentrifugal determination of the molecular weight of polysaccharide B-1459 revealed that molecular aggregation is the cause of difficulty in applying this method.

Toxicity of B-1459 was investigated in the Pharmacology Laboratory of the Western Division. No unfavorable effects were observed. Safety evaluation data were obtained with rats and dogs for inclusion in industrial petitions to Food and Drug Administration pertaining to a proposed use of the polysaccharide as a food-thickening agent. Approval for food use would open new, large outlets for B-1459.

5. Fermentation acids. In studies under a PL 480 grant to the University of Lodz, Lodz, Poland, strains of *Aspergilli* were mutated to increase yields of itaconic acid, but no strains capable of direct conversion of glucose to itatartaric acid were reported. However, the conversion of itaconic acid to itatartaric acid is a simple step that can be accomplished by an oxidative dehydratase.

Other research on fermentation acids, recently undertaken under PL 480 grants to the University of Tokyo, Tokyo, Japan, includes studies on production of D-tartaric acid and investigations on formation of mevalonic acid. No significant results have yet been reported.

### C. Biological Pesticides

1. Biological insecticides for Japanese beetle. Substances that might be expected to preserve cell wall integrity, such as trypsin inhibitor, nucleic acid and acetyl choline, have increased sporulation of *B. popilliae* on solid culture media. Content of borate in the medium was found to influence sporulation significantly. Increased borate levels stabilized cells but decreased sporulation presumably by decreasing cell wall permeability. A technique for the isolation of whole cell walls of *B. popilliae* has been devised. Free spores of this organism were also isolated and characterized. Extensive experiments indicated that *B. popilliae* cultures do not carry a demonstrable phage even though minute lytic areas have sometimes been observed in test plates inoculated with ultraviolet-irradiated *B. popilliae*.

Additional information was accumulated on the composition of hemolymph from healthy and diseased grubs and on improved media and methods for increasing cell populations of cultures. Grubs have been successfully obtained by breeding of adult Japanese beetles in the laboratory.

Contract research on artificial culture of milky disease bacteria has been completed at Michigan State University. Media and conditions for producing cell populations up to 3 billion per ml. (biphasic) and viability up to 14 days were developed. An unexplained need for barbituric acid in synthetic media was discovered. A sporulation medium containing this ingredient caused 50-100 percent of the cells to develop sporelike bodies. Other important findings point to the possible importance in sporulation of acetate metabolism and intracellular accumulation of hydrogen peroxide. Other contract studies at Michigan State University revealed that ribosidase activity was greater in in vitro "spore-bodies" than in vegetative cells, but no activity was observed in spores developed in diseased grubs. Up to 78 percent of



"sporelike" bodies were obtained by addition of  $\beta$ -hydroxybutyrate to cultures on the third day of incubation. Supplemental feeding of glucose and L-arginine to the medium resulted in total cell populations of  $4 \times 10^9$ /ml. at 48 hours of incubation and 80 percent "sporulation" by the 16th day of incubation.

Studies on the sporulation factor at the University of Illinois showed that addition of a sporulation factor isolated from several strains of B. cereus, popilliae and lentimorbus to a growing culture of B. popilliae damaged the cells but did not induce sporulation. This uncharacterized sporulation factor was found both in B. popilliae strains that fail to sporulate on solid medium and in the strain that does. These observations indicate that failure to sporulate is due to some sort of cell malfunction but not to absence of any recognized sporulation factor. The sporulation factor, now named "sporogen," has been isolated in pure crystalline form and partially characterized. Only 19 mg. was recovered from 20 pounds of B. cereus cells.

Scientists at the University of Minnesota have produced and isolated auxotrophic mutants of B. cereus T which will serve as a source of transforming DNA in transfer experiments.

At Baylor University College of Medicine, electron microscopy of thin sections of B. popilliae cells showed a 3-layered cell wall, which is unique among gram-positive bacteria, and has provided other extremely detailed information on the fine structure of spores of B. popilliae. Fluorescence microscopy showed that old cells of B. popilliae experience leakage of cell contents.

Studies on stabilization of vegetative cells of B. popilliae are in progress at Kansas State University. Some success has been achieved in devising systems that prolong cell life as judged by plating methods. However, no system has yet proved effective for preservation of cells in soil.

The success in achieving high percentages of "sporelike" bodies suggests that the sporulation problem may now be beginning to yield to the massive scientific effort applied against it. Of first importance, however, is to determine if these "sporelike" bodies are in fact spores and, if not, what their relationship is to true spores. The characterization studies on spores at the Northern Division and the electron microscopic studies at Baylor University provide the basis for differentiating true from imperfect spores.

2. Plant antibiotics. An additional 27 streptomycetes were found that yielded promising nonpolyenic antifungal activity. Trials of 20 of the most promising antibiotics against 12 plant diseases and 1 nematode infestation are now in progress at Boyce Thompson Institute. These trials are being conducted on host plants so that the results when available should provide definitive indication of the value of the antibiotics tested.



#### D. Feed and Food Products

1. Microbial carotenoids. In runs in 20-liter fermentors, a maximum wet-cell weight of 19 g. per 100 ml. of medium has been obtained with Chlorella "vulgaris". This organism produces xanthophylls, mainly lutein and zeaxanthin. The present wet-cell weight is equivalent to 3-6 mg. of xanthophyll (dry basis) per 100 ml. of medium. A tenfold increase in yield would, however, be needed to have a process justifying industrial interest. Ten pounds of a culture of another promising alga, Chlorella variegatus, was produced in 60-liter fermentors for more detailed study. At glucose concentrations of 3 to 10 percent, chlorophyll formation is inhibited and xanthophylls (about 75 percent lutein) are the predominate product. Resting cells of some 200 molds, yeasts and bacteria failed to convert  $\beta$ -carotene to xanthophyll.

Contract research by the Arthur D. Little Company on stabilizing fermentation  $\beta$ -carotene showed that the half-life toward oxidation could be extended many months by embedding the  $\beta$ -carotene in gelatin containing a carbohydrate such as dextrose. A polymer precipitation technique has provided limited success, but conventional coating techniques have so far been unsuccessful.

Contract research by Consolidated Laboratories, Inc., on use of antimetabolites to develop high-yielding strains of the  $\beta$ -carotene-producing microorganisms has been initiated but no results have been reported.

2. Fermented wheat foods. Tempeh fermentations with wheat as substrate were found to be more susceptible to contamination by undesired microorganisms than those based on rice or soybeans. Present results suggest that fermentation at low pH's may prove to be an effective means of minimizing bacterial contamination. At the same time, low pH may provide the further advantage of favoring a better or more complete fermentation.

Partially purified proteolytic enzymes produced by Rhizopus oligosporus NRRL 2710 (the organism used for making tempeh from wheat) was found to be 3 to 5 times more potent than pepsin. Activity was particularly high toward wheat gluten. Study of enzymes involved in production of fermented wheat food products is providing new information that should have application in developing such foods and that has important implications in relation to enzyme chemistry and technology.

3. Vitamin B<sub>13</sub>. In research under a PL 480 grant at the "Giuliana Ronzoni" Scientific Institute of Chemistry and Biochemistry, Milan, Italy, fractions have been isolated from dried distillers' solubles comprising peptidic substances, at least seven phenols, carboxylic acids and carbonyl compounds. Characterization of the fractions is in progress. No single component isolated thus far produces a growth response in test animals like that obtained with concentrates of distillers' solubles.

4. Improved feeds by mutation of yeasts. Under a PL 480 grant to the National Institute of Agronomic Research, Paris, France, studies are in progress that have as their objective selection and mutation to obtain strains of yeast capable of producing high quantities of sulfur-containing amino acids for use in improvement of cereal-based feeds deficient in these amino acids. Mutant strains of the yeasts Candida utilis and C. tropicalis were obtained from cells resistant to a mutating agent, ethionine. Cells of the C. utilis mutants revealed a 10-percent net increase in intra- and extracellular methionine. There was negligible increase in the cystine content of the cells, although these and all other mutated C. utilis cells contained more nitrogen than the parent strains. The increases in nitrogen and particularly in methionine are considered significant.

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WHEAT AND BARLEY - FOOD AND FEED PRODUCTS AND PROCESSING  
Western Utilization Research and Development Division, ARS

Problem. The dominant factor in the wheat economy of the United States continues to be that production capacity has outpaced even the substantially expanded foreign markets of recent years. Supply and demand have been brought closer to balance only through production curtailment and substantially subsidized foreign marketing. The resulting tendency to depress the agricultural economy has an important influence on the entire national economy. Despite the shrinkage in numbers of the farm population in the past 20 years, agriculture remains the principal ultimate customer of our nation's industrial products and services. Agriculture also provides the largest share of our commercial exports and contributions to a favorable trade balance. Wheat is a major commodity in both our domestic and foreign trade considerations, and is a primary segment of our agricultural economy. Sustained further gains in wheat markets are necessary to ease restrictions in production and to strengthen trade balances. Especially needed are increased domestic usage and commercial exports. Domestic usage of wheat has been relatively constant for many years, but encouraging signs of a stabilized per capita food consumption and a trend for greater feed usage in the past two years suggest that total domestic usage can soon begin to exceed the prevailing 600 million bushel total. For the past several years exports have surpassed domestic usage, but a large proportion of the exports have been concessional sales and donations with long-range market-building objectives. Increased world supplies of wheat and restrictive political decisions in the European Economic Community have contributed to seriously reduced commercial exports during the past marketing year. Everything possible must be done to increase total wheat markets, but especially those in which payments are made in dollars. The export donations and concessional sales in excess of 500 million bushels provide food where it is most needed in the world and serve immediate Defense and State Department missions, as well as the long-range market development for U.S. agriculture. The less than 200 million bushel commercial export of wheat, however, is the most important segment of the market to be increased. New processes to elicit maximum quality performance of wheats and flours in national products of all markets will help significantly in this regard. A more efficient conversion of wheat, mill feeds, and barley to meat will offer an improved opportunity to use more of the grains, although at a lower return to growers than from food products. New wheat food products specifically adapted to conditions of use in every region of the world will help materially in popularizing this valuable food grain in areas where it is now virtually unknown. Development of simplified methods to process the products at point of use will speed adoption of the many ways wheat can be used as food. An essential foundation for a successful product and process development program is a strong program of basic research on the composition of all classes of wheat and the fundamental properties of their constituents.



## USDA AND COOPERATIVE PROGRAM

Research on utilization of wheat and barley for food and feed seeks to solve the most urgent problems hindering the development of markets for the full productive capacity of U.S. agriculture. The emphasis is on (1) expansion of overseas dollar markets for U.S. wheats; (2) development of new wheat food products for long-term market development in food-short nations abroad; (3) raising the domestic consumption of wheat foods by increased variety, quality, and convenience; and (4) finding means to upgrade wheat millfeeds and barley so they will be more valuable as livestock and poultry feeds. Basic research on the fundamental chemical and physical properties of wheat and barley constituents and on the functional properties of wheat flour constituents supports the product development and problem-solving segments of the program.

The long-standing and widespread popularity of wheat as a food substance depends chiefly upon the unique properties of its proteins, but adequate understanding of how these proteins interact with one another and with the other important flour constituents to bring about desired properties in wheat foods has not yet been achieved. Despite centuries of skillful empirical use of wheat in milling and baking, quality problems remain to impair the efficiency of using this valuable food material, particularly as changes in technology come along in the processing industries. Product development likewise is slowed by lack of adequate fundamental information on properties of components. Basic chemical studies are therefore conducted on wheat proteins, lipids, carbohydrates, and enzymes and on specific interactions among these components. Chemical changes that occur during the artificial maturing of flours or that occur during the mixing of doughs are studied to learn how wheats differ from one another, how each should best be processed, and how inherent quality variations can best be accommodated in desirably standardized and mechanized processing. Such considerations are also very important for our dollar wheat markets overseas because in the more important, such as Japan and Western Europe, highly mechanized production of bakery products is increasing as it has in the U.S. New methods for the artificial maturing of hard red winter wheat flours are especially needed for the European markets because methods commonly used elsewhere are not allowed and because as strong a baking performance as possible is needed to meet the competition from the high-quality wheats marketed from Canada. Our search for new maturing methods requires an understanding of the complex mechanisms of the process which involve enzymes, sulfhydryl groups on proteins, unsaturated fatty acids, and atmospheric oxygen, among other factors. Fundamental work of this type also aids in the development of new wheat food products being designed for varying needs in specific foreign countries. Development is well along, for example, in the enzymatic conversion to soluble form of the protein of wheat and wheat materials for high-protein beverage products suitable for infant feeding abroad. Other work involves development of high-protein formulations resembling meat products in flavor and texture, again for markets overseas where protein supplies are scarce.

Research is also continuing on development of inexpensive, easily processed types of products related to bulgur, such as WURLD wheat and malted products to help meet both calorie and protein needs in developing countries and to help build future dollar markets for U.S. wheats.

Several lines of work are primarily concerned with improving the quality and variety of wheat foods for the high-return domestic market. Such work includes a continuing study of the mysteries of the flavor and aroma of freshly baked bread so that product quality can be kept up to par or improved as economic factors force changes in baking and distribution methods. Related work seeks to overcome the inability of many flours to carry the amount of nonfat dry milk customarily and desirably used in breads when they are produced by the increasingly important continuous-mix methods. Work is being started on the fermentation aspects of bread production, not only to improve control of flavor production, but also to develop frozen doughs of better quality and stability. Several types of snack and convenience foods based on WURLD wheat and malted products are being developed to broaden the base of wheat food offerings to consumers.

Increased attention is being devoted to better utilization of the large tonnages of feed products from flour milling now marketed at income depressing prices. Methods are under development to recover the high biological value protein from millfeeds for use in food products and to make energy components more easily utilizable by poultry and livestock. Factors affecting the biological availabilities of protein and energy constituents of wheat feeds are being measured to assist their attainment of a better competitive position in modern feed processing based on linear programming and computerized formulations.

Research is conducted by the Western Utilization Research and Development Division at Albany, California; under contracts and grants at Pullman, Washington; Chicago, Illinois; Manhattan, Kansas; Madison, Wisconsin; St. Paul, Minnesota; Menlo Park, California; and Corvallis, Oregon; and under P.L. 480 grants in England, France, Poland, Italy, Australia, Switzerland, and Israel.

The Federal program of research in this area totals 36.7 professional man-years, including one scientist whose salary is provided by the Farmers Co-Operative Commission Company under a Memorandum of Understanding and 10 contracts and grants providing research at a rate of approximately 8.6 professional man-years per year. Of this number, 20.0 are assigned to investigations on chemical composition and physical properties; 14.5 on new and improved food products and processing technology; and 2.2 on new and improved feed products and processing technology. In addition, the Division sponsors 15 research grants under Public Law 480 including 13 on basic studies and 2 on applications of research.



## PROGRAM OF STATE EXPERIMENT STATIONS

State stations conduct a comprehensive program of basic and applied research directed to increasing or improving the utilization of wheat and barley. These studies involve determining the influence of environmental, agronomic, harvesting and storage factors on the ultimate milling and baking quality of new selections and established varieties. Physical dough properties and baking characteristics are evaluated to provide guidance to cereal breeding programs and full knowledge of quality to the milling and baking industry. Testing often begins with laboratory micro-quality tests and extends through full-scale milling treatment.

The newer types of malting barley are studied to determine adaptability for malting. Basic composition, including enzyme content, is followed and these properties are related to potential for commercial malting.

The unique value of wheat flour for baking is due to special characteristics of the proteins. By labeling these proteins with radioactivity, the changes which take place during the various steps of baking are evaluated. Powerful new electrophoretic methods are being used to show that flour contains many proteins rather than the three or four previously thought present. Analytical techniques are being developed to determine quantitatively the protein content of wheat flour samples. These can also be used to develop wheat varieties containing more of the desirable proteins. Development of higher protein quality would improve the value of wheat in the diets of underdeveloped countries. For example, detailed research on the amino acid content of wheat, especially lysine, provides information of wide interest from a nutrition standpoint.

A number of basic studies are being carried out to further elucidate fundamental principles involved in conversion of cereals into food products. These include determination of physical grain properties, physical properties of small particles important in flour milling technology, and the nature of the enzymes, lipids and water soluble gluten fractions. Hydrogen bonding in proteins is being studied to clarify its role in determining the baking quality of wheat proteins.

The aroma and flavor of fresh bread and bakery products have universal appeal. Researches directed to determining the constituents responsible for the odor and flavor of bread and bakery products continue. Modern techniques such as gas chromatography are being used in these studies.

As a part of a broad study on conditioning of wheat, the changes in some properties of the aleurone cell layer caused by steam-conditioning are being investigated. Present data indicate denaturation of the proteins of the aleurone cell layer may be a major change brought about by steam-conditioning.

Although application of bleaching and improving agents to flours has been practiced for many years by the industry, the precise role played by chlorine



in the bleaching process has not been elucidated. Researchers are applying the tool of x-ray fluorescence spectroscopy for the analysis of the chlorine content of unbleached and bleached flours and their major biochemical fractions. The mechanism of improver action in cake flours is also being studied by determining the relation between flour specific surface and chlorine distribution. Other studies involve problems of air classification, the preparation of fine and intermediate fractions of wheat flour and the structure of the starchy endosperm.

Attempts to develop new and improved cereal products are directed to determining the fundamental properties of doughs and the effects of freezing upon them. Stability tests and related investigations on a wheat wafer for shelter rations are being conducted in cooperation with USDA (WU,ARS). Survival rations, including cereal products, are being evaluated.

Economic feasibility studies give attention to use patterns of barley and wheat of differing quality and the supply and flow patterns of wheat.

The total research effort on wheat and barley utilization research is approximately 12.2 professional man years.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Chemical Composition and Physical Properties

1. Proteins of Wheat. Protein quantity and quality, more than any other compositional factors, determine the bread-making and many other baking qualities of wheat flour. Mixing and kneading transform flour paste into strong, smooth, resilient dough. The protein of the flour determines whether or not the dough will be elastic enough to retain its bubbles of CO<sub>2</sub> as the yeast ferments and at the same time be fluid enough for the bubbles to expand and form a bold, attractive loaf. The spread of cookies, the formation of cake crumb, and the texture of crackers also depend considerably upon the amount and character of wheat protein although each demands differences in these factors from those ideal for bread making.

Bonds are made and broken within and between protein molecules as bread dough is worked. Non-protein wheat constituents and other dough ingredients affect the making and breaking of protein bonds. The rate and extent of all the reactions determine the quality, shape and size of the baked loaf. The association, interaction, or aggregation of flour proteins and the significance of such behavior to the functional properties of doughs and batters comprise an important area of utilization research. Increased knowledge of these factors is necessary for significant new advancements in product improvement, development of new products, and greater usefulness of U.S. wheats in foreign dollar markets.

A new concept emanating from our research indicates that protein particles must disaggregate during mixing before an appreciable protein network or

film can form. Such a network or film is widely regarded as the essential role of wheat protein in the development of strong bread doughs. Differences in the size and stability of protein aggregates of flour prior to mixing may explain the wide differences among flours as to their dough-mixing requirements. In order to test our ideas in this area, we have separated small amounts of wheat flour components and groups of components that can be used for baking or other tests in reconstituted doughs or simpler systems.

When a strong, high-quality flour is treated with dilute acetic acid, the residue is a water-insoluble protein-carbohydrate mixture in the form of a highly hydrated gelatinous mass. If the same extraction procedure is applied to doughs, the amount of this residue is less, and it decreases rapidly with additional mixing. The insoluble material, which consists of a protein that is strongly associated with hexosan and pentosan materials, appears to be a major factor governing the mixing requirements of flours, and possibly their baking performance. The increasing solubility that accompanies mixing is probably related to separation of the associated carbohydrate and protein.

Gluten is the major protein substance that provides flour with the functional characteristics associated with good baking quality. We are investigating the effects of both crude and purified gluten preparations on the behavior of doughs. The proportion of aggregated protein in the gluten preparations was lowered by dispersing them in dilute acetic acid in a blender or by centrifuging them to remove aggregates. Doughs containing crude glutes required the longest mixing times to reach maximum resistance. Doughs containing gluten subjected to the dispersing and centrifuging treatments required much less time. These results are consistent with the assumption that the protein aggregates present in flour must be disaggregated in the dough-mixing process before film formation can occur.

Gluten can be separated into glutenin and gliadin. Gliadin components are relatively small molecules (molecular weight about 25,000) and thus cannot be expected to contribute significantly to viscoelastic properties of doughs unless they associate with other molecules. Research is conducted on the effect of various conditions on gliadin-protein associations, particularly under conditions similar to those in bread doughs. Some gliadin components have been isolated and their properties are under preliminary investigation.

Basic studies on the types of bonding responsible for cohesiveness and related physical properties of gluten were concluded in contract research at the Midwest Research Institute in Kansas City, Missouri. They developed procedures for preparing freeze-dried crude gluten, purified gluten, and glutenin of high-quality in quantities of several hundred grams each, and they studied the viscoelastic properties of the preparations under various chemical conditions. When the water content of all the gluten systems was increased, the stress strain modulus decreased. Viscoelastic properties of crude gluten were not significantly changed by variations in salt concentration, urea concentration, or pH. Glutenin has a higher elastic modulus and



slower rate of stress decay than has crude gluten. Complete acetylation of free amino groups destroyed the cohesiveness of gluten, indicative of the role of hydrogen bonding in determining gluten's cohesive properties. Methylation of free carboxyl groups appeared to reduce the strength of crude gluten relatively more than that of purified gluten or glutenin. However, the effect might have been related to starch impurities in the crude gluten. Blocking of free sulfhydryl groups reduced cohesion of the purified gluten, but it did not affect the cohesion of crude gluten or glutenin.

Flour proteins, yeast, and other bread ingredients are rich sources of enzymes. During dough mixing and fermentation, enzymes become increasingly active, causing chemical breakdowns and recombinations that affect bread quality. Protein-splitting enzymes of wheat flour, for example, were found to hydrolyze the  $\alpha$ -casein component of nonfat milk solids. Data from experiments at different pH levels indicate that the same system(s) of flour proteinases are active on hemoglobin and  $\alpha$ -casein substrates. When  $\alpha$ -casein was reacted with wheat flour, three characteristic hydrolysis products resulted. They were electrophoretically identical with reaction products obtained when a protease concentrate from flour was used instead of whole flour. The isolation of these specific protein fractions is further evidence that the protein-splitting activity of wheat flour is of a restricted and specific nature. The splitting of milk protein by wheat flour enzymes may be connected with the differential tolerance that flours have for added nonfat milk solids in dough formulas for continuous-mix bread making.

Two major types of protein-splitting enzymes of flour are distinguishable by their solubility characteristics. They differ in their activity dependence on pH and ease of denaturation. One has a maximum activity at pH 4.5 and is inactivated if sulfhydryl groups are blocked by use of a chemical reagent. This enzyme is relatively insoluble in water. Earlier attempts to extract it from flour generally led to its inactivation. We were able to extract much of the proteinase from flour using dilute acetic acid. With four sequential extractions, some 70% of the total protein-splitting activity was obtained, and most of the rest of the original activity could be accounted for in the residue. The extraction of this enzyme without loss of activity opens the door to its further purification and characterization so that studies can be conducted to determine its role in flour protein interactions. The other enzyme appears to have a maximum activity at about pH 3.8. It is also being characterized.

Basic chemical information on the function of sulfhydryl groups in protein-splitting enzymes from wheat is being developed at the University of Poznan in Poland under a P.L. 480 research grant. To determine more specifically the dependence upon sulfhydryl groups for protein-splitting properties, one of the wheat enzymes was isolated and purified and its homogeneity established by paper electrophoresis. This enzyme (designated protease A) contains both reactive and masked sulfhydryl groups. Its amino acid composition is unusual in that the proportion of cystine is high. Gluten, when incubated



with the isolated wheat proteases, is relatively resistant to their hydrolytic action, compared with other standard proteins such as hemoglobin.

2. Dough Rheology. Control of the mixing processes in large bakeries depends upon empirical measurements of resistance to mixing and extensibility of mixed doughs. In order to obtain fundamental data that will be useful in developing more sophisticated equipment and procedures for the testing of flour for bread-making quality and for making significant improvements in flour usage, we have initiated a series of basic investigations. In contract research recently started at Stanford Research Institute in Menlo Park, rheological properties (physical factors concerned with the plastic flow of materials) of wheat flour dough are being studied to determine relationships between molecular organization in doughs and their baking characteristics. Additional basic investigations are being supported by Public Law 480 grants to the Rheological Laboratory of the Israel Institute of Technology at Haifa and to the Bread Research Institute of Australia in North Ryde, New South Wales. Two different approaches are being taken under these research grants, but both are directed toward providing better measurements of the viscoelastic properties of flour doughs.

The project in Israel provides rapid measurements of tensile stress-and-strain relationships, relaxation time, and elastic recovery behavior in dough specimens taken at intervals during a dough-mixing cycle. These very precise measurements revealed that during an extensograph run of about 20 seconds the apparent coefficient of viscosity of doughs decays in a dramatic way. A 20-second extension of a molded piece of dough reduced its viscosity to less than 1/5000. Without the extensograph treatment, the viscosity of molded dough held up to 45 minutes. Salt had little or no influence on the viscous behavior of dough, and hence the pronounced influence of salt on extensograph values must be ascribed to something other than an effect on viscosity.

Studies such as these are complicated by the intricate interrelations of flow properties of a dough and the continuous changes, both physical and chemical, that occur in raw material used over a number of weeks or months. Stored flour changes, so that a series of experiments may reflect changes in the raw material in the system. Considerable effort must be taken to be certain that experiments are not unduly affected.

The scientists at the Bread Research Institute of Australia are using equipment designed to measure viscoelastic properties by applying a dynamic sinusoidal loading in contrast to the measurements employing a static loading in the Israel project. The two projects thus complement one another. The research grant in Australia is recent, and only preliminary findings have been reported thus far. They indicate that the equipment as now designed will be satisfactory over a low range in the frequency of stress application. The possibility of extending the usable range of the equipment by modifying the equipment design will be investigated.

3. Analysis of Protein and Related Components. Because the development of bread dough and the structure of wheat bread depends on the particular

proteins that wheat contains, we seek basic knowledge of these proteins in order to control raw materials and improve products and processing technologies. To understand wheat proteins, the individual proteins must be separated and their chemical composition and functional properties studied. Classical separation of proteins is based on differences in their solubilities in various liquids and solutions. By the most recent laboratory techniques, soluble proteins are studied by gel electrophoresis, which involves differences in molecular size and charge. Once separated by such techniques as gel filtration or preparative electrophoresis, the proteins are characterized by molecular weight, amino acid composition, and biological activity, particularly their functions as enzymes.

An important property of all proteins is related to their antigen-antibody reaction in the blood stream of animals. Specific antibodies develop when a foreign protein enters the blood. Upon subsequent exposure to the specific protein, these antibodies react, causing allergies or anaphylactic shock in animals or, in the laboratory, precipitation of blood components. This antigenicity, which can serve to characterize proteins, is the basis of the immunoelectrophoresis technique innovated by Dr. Grabar at the Pasteur Institute in Paris. With grant funds under P.L. 480, Dr. Grabar is continuing his research on cereal proteins. He concluded that an antigenic identity exists between alpha-amylases of barley, rye, and wheat, and that partial identity exists between these alpha-amylases and those of maize and oats, even though the latter are of different electrophoretic mobility. The beta-amylases of barley have antigenic identity, even though their molecular sizes differ. The presence of identical antigenic sites on protein molecules from different cereal sources may provide useful clues for ultimate studies of the molecular architecture of such proteins.

Immunochemical studies of soluble proteins from flour are also conducted in-house, including comparisons of hard red spring and durum wheat varieties. The results obtained so far by conventional and immunochemical gel electrophoresis techniques indicate that no clear qualitative differences exist between the soluble proteins of durum and hard red spring wheats. Any differences in processing quality related to the soluble proteins would thus appear to be an influence of quantity of such components, not kind. Confirmation of these results depends somewhat upon obtaining larger quantities of isolated soluble protein components. Work has been initiated on such a separation by use of continuous preparative electrophoresis on free-film equipment.

In contract research at Washington State University in Pullman, Washington, scientists are developing techniques necessary to use radiotracers in wheat proteins in order to detect the participation of individual proteins in the changes that occur during mixing and baking of bread. Wheats were grown in chambers under conditions whereby radioactive carbon atoms were incorporated in the various proteins of the wheat produced. Physical and chemical methods will be used to follow changes in these proteins after the wheats are milled to flour and the flour mixed into dough and made into bread in subsequent



studies. Slight differences in growth chamber conditions and field conditions were encountered that appear to be influencing the protein composition of wheat being grown in this investigation. This observation will be scrutinized with great interest because it implies a control of wheat protein composition by cultural conditions of production.

Basic studies on the solubility of wheat gluten proteins in aqueous systems were initiated with a P.L. 480 grant to the National Center for Scientific Research at Montpellier in France. Although gluten is considered insoluble in water, prolonged extraction after removal of the easily soluble protein provides additional quantities of a gliadin-like protein material. Attempts will be made to determine any correlation between quantities of this wheat component and baking quality of flour.

Another attempt to study insoluble gluten involves application of ultrasonic vibrations to suspensions of wheat gluten for the purpose of fractionating and modifying individual constituents of the protein for further identification. This work, supported by a P.L. 480 grant, is being conducted at the National Institute for Agronomic Research in Paris, France. Greater changes were induced by ultrasonic vibrations in gluten dispersed in acetic acid solutions than could be obtained using undispersed gum gluten. Solubilized material is being fractionated on dextran columns and the fractions are characterized by electrophoresis methods. Thin-layer chromatography of component amino acids is also being used to identify separated proteins and protein fragments.

At St. Albans, England, a P.L. 480 grant has been made to the Research Association of British Flour Millers, where protein is being solubilized by a novel method. Reactions with metallic and acidic ions, such as cupric and sulfite, were previously found to solubilize wool and feather proteins. Similar reactions are being adapted to study the less soluble protein components of wheat. Gluten solubilization has been obtained by use of sulfite ions in solution but good characterization of components by gel electrophoresis has not been achieved. Behavior on Sephadex columns is being interpreted in order to characterize different components of the gluten solubilized by sulfite treatment.

The interactions of proteins with ions and small molecules often play a basic role in their function. Such interactions can be examined by measuring molecular volume changes and such measurements are under investigation using the interactions of calcium and magnesium ions and adenosine triphosphate as a model system. In other studies on thermal decomposition of proteins, mass spectrometric data indicate that gliadin and glutenin undergo little degradation in the dry state up to high temperatures (170° C.). At higher temperatures, sulfur-containing amino acids break down first, with further breakdown and charring at temperatures above 200° C.

Because polysaccharide components of wheat are known to be related in some way to the baking quality of flours, we are studying the polysaccharides and



the related glycoproteins of wheat flour. Procedures for extracting water-insoluble pentosans from wheat flour were developed and a number of fractions separated by chromatographic procedures with various solvent combinations. While some separated fractions differ in chemical reactivity when hydrolyzed, all fractions examined so far contain the same groups of 5-carbon and 6-carbon sugars after hydrolysis. Unlike water-soluble pentosans that have been studied elsewhere, these water-insoluble ones have not contained galactose, and they have a very low protein content. We have found that water-insoluble pentosans can be solubilized in fluorinated organic solvents without the side reactions that occur between pentosans and some organic solvents.

The pentosans that exist in combination with proteins as glycoproteins are involved in one of the oxidation phenomena noted in wheat components. Such pentosans form gels when they are oxidized with bromate and iodate. The changes that occur during oxidation of pentosan glycoproteins are being studied at the Swiss Federal Institute of Technology at Zurich under a recent P.L. 480 grant.

At the University of Bologna in Italy a basic investigation of nitrogenous components of wheat germ is being supported by P.L. 480 funds. Wheat germ contains nitrogenous components of high biological value and a rational utilization of wheat germ in connection with nutrition, whether for food or feed, depends upon a better knowledge of these components than we now have. These studies have revealed the presence in wheat germ of appreciable quantities of protamines, several of which had not previously been found in plant sources. These include compounds found in putrefying animal flesh and others that are involved in animal reproduction.

4. Maturation of Wheat Flour for Bread Making. The addition of oxidative improvers to wheat flours in modern milling and baking eliminates the lengthy natural period of maturation required for some types of flour, particularly those made from hard red winter wheat. Effective treatments have been developed that are widely regarded as safe. Even so, regulations of continental European countries prohibit the treatment of food materials with almost any chemical agent, so that flour-maturing agents common in America and the United Kingdom are not allowed. These regulations hinder the development of commercial markets for our most abundant wheats. Therefore, our research program includes a number of projects aimed at achieving a better understanding of the maturation process with the long-range objective of finding acceptable alternative methods for maturing wheat flour.

Evidence is accumulating that flour lipids are involved in wheat flour maturation. We have found that the ratio of non-polar to polar lipid is higher in the hard red spring wheat flour, which requires little or no maturation, than in hard red winter wheat flour, which requires maturation or oxidative improvement. Treatment of the flour with oxidants affects the ratio. Preliminary data indicate that the ratio is slightly increased by bleaching with benzoyl peroxide or acetone peroxide. These studies will continue.

During prolonged mixing of dough, its sulfhydryl content increased if oxidation was prevented by blanketing the mixer system with nitrogen. In the first five minutes of mixing, sulfhydryl content decreased rapidly, then returned rapidly to the original level and increased with subsequent mixing. If lipids were extracted from the flour with carbon tetrachloride, the initial rapid loss of sulfhydryl during dough mixing was eliminated. This observation implicates lipids, probably oxygenated lipids, in some way with the changes in sulfhydryl.

Other investigators have reported that bromate and iodate oxidative improvers are consumed at a steady rate during dough mixing, but at a much slower rate when the dough is resting. They proposed that the effect of mixing was to diffuse the oxidants and thus speed the rate of reaction with sulfhydryl groups. Our findings support a different concept: that mixing releases sulfhydryl groups making them available to react with the oxidants. We attribute the initial loss of sulfhydryl groups (when dough was blanketed with nitrogen) to the oxidation of the few reactive groups originally present in the flour. Subsequent reactions depend upon the release of sulfhydryl groups by mixing.

Grant research is in progress at the University of Wisconsin to determine the important oxidative enzymes in wheat flour and the nature, extent, and conditions of their activity in dough systems. Lipoxidase, catalase, and cytochrome oxidase activities were observed in extracts of whole wheat and endosperm, and ascorbic acid oxidase and pyrogallol oxidase in whole wheat but not in endosperm. Much higher levels of enzyme activity were found in mill fractions, break shorts, reduction shorts, bran, and red dog than in flour. Of the enzymes studied, lipoxidase and catalase were by far the most active in the one hard red winter wheat variety studied so far. Other varieties will be examined, and quantitative estimates of enzyme activity will be made.

Studies of the coenzyme role of riboflavin of wheat endosperm are conducted under P.L. 480 at the Agricultural Higher School in Poznan, Poland. Flavin nucleotides are the indispensable components of several enzymes involved in biological oxidation. The amount and form of flavins in wheat flour from the various market classes of wheat and any coenzyme function of commercial importance are being determined. Appropriate methods are being developed for extraction of both loosely bound and tightly bound flavins from wheat flour for accurate quantitative determination of the flavins. The general procedure for flavin concentration by phenol had to be rejected as inappropriate for flour extracts, but concentration by cation absorption appears to be of value. Development of analytical procedures and selected instrumentation were tested and advanced to a routine basis.

5. Lipids and Lipoproteins. Closely related to the studies of wheat flour maturation are compositional investigation of lipids and lipoproteins. Benzene-soluble material from wheat flour was precipitated with acetone to give a material that contains lipoprotein, and the nature of the chemical



binding between the lipid and the protein was investigated. Since phospholipids are good metal chelaters, the first approach was to study the metal binding of phospholipids. In model compounds made by removal of fatty acid radical groups from two typical phospholipids, phosphatidyl serine and triphosphoinositide, the stability of metal complexes was determined by pH titration in aqueous micellar dispersions. Stability constants for the intact lipid metal complexes were 10 to 100 times greater than those of deacylated models. This greater stability is attributed to electrostatic properties leading to micellar aggregation in aqueous solvents. Surface charge characteristics greatly favor complex formation. Our hypothesis that lipids and proteins can interact through mixed metal chelate formation is supported by these data.

In contract research conducted at Kansas State University, lipids were extracted from flour milled from 16 wheat varieties (including hard red winter, hard red spring, soft red winter, durum, and soft white club wheats) for comparison of lipid composition with baking characteristics. Differences in lipid composition were relatively small, and the relationship of the lipids to baking characteristics of the flours was not apparent. These flours represent substantial quantitative differences in protein content and a wide range of responsiveness to oxidative improvers as well as substantial differences in baking quality. Interrelationships of these characteristics have not yet been established.

A basic study, supported by a P.L. 480 grant, of the lipids of whole wheat, ground wheat flour, bran, and other milling fractions is being conducted at the French School of Milling in Paris. The fatty acid composition of wheat lipids is being compared for a number of market classes of United States wheats. In addition, the effects of wheat lipid components on dough mixing characteristics are being investigated. Total lipids were extracted from flours and were fractionated. Defatting increased the Alveograph resistance to extension of doughs but after reaching its maximum, the resistance decreased faster than for normal doughs. If oxidation was allowed, the decrease in resistance was even faster. If sulfhydryl groups were blocked and prevented from reacting in the dough, there was no rapid loss of resistance. Reincorporation of unsaturated (oxidizable) fatty acids increased the resistance. These studies are providing important clues to the mystery of bread dough quality involving protein interactions, lipids, and oxidative improvers.

Flours and glutens from 10 United States wheats, including club, soft, hard, and durum samples, are being studied under a Public Law 480 grant to the National Institute of Agronomic Research in Paris, for their content and type of phosphorus compounds related to lipid and protein composition. The phosphorus in albumin-type proteins is generally of a phytic nature, whereas that in globulins is in the nucleic acid form, chiefly desoxyribonucleic acid. Hard spring and durum flours contain more phytic acid phosphorus than does hard winter wheat flour, and soft wheats contain less phospholipidic phosphorus and less gluten ribonucleic acid phosphorus than do hard wheats



and durumms. Methods of analysis are still being refined in this research and it remains to be seen whether measurements of phosphorus components will be useful to predetermine baking quality of various wheat flours.

Under a P.L. 480 grant, the National Institute for Agronomic Research in Paris, France, is investigating the lipase activity of wheat as a function of water vapor tension, partial pressure of oxygen, and temperature. Methods are being developed for extracting and isolating lipase of wheat and determining fatty acids released by lipase action in wheat held at very low moisture contents. The lipase activity of wheat at various stages of germination and growth has also been studied. Lipase activity appeared to be closely correlated with the growth and size of wheat seedlings. Temperature conditions that retarded or enhanced plant growth also retarded or enhanced the lipase activity.

6. Flavor. Fresh-bread flavor is an important factor in enhancing bread consumption. The instability of fresh-bread flavor remains a technical challenge to research. Basic investigations are revealing the chemistry associated with bread flavor. Now if we can learn specifically what components are responsible for flavor and how to measure them, we can provide the cereal chemist and baker with tools for flavor enhancement and flavor stabilization. An aroma concentrate resembling bread flavor was made from a brew of normal bread preferment ingredients except that flour was omitted. An extract was taken after four hours of fermentation and concentrated. This concentrate had a vaguely reminiscent odor of bread, but when it was heated in air at 210° F. it produced a very definite aroma of baking bread. Although the extract as obtained is a complex acidic liquid, neutralization of the acid did not affect aroma characteristics. Gas chromatography analysis indicated that the concentrate extracted from preferments had many of the same components as did condensates of baking oven vapors, except that the oven vapors had several very high-boiling components not present in significant quantity in the preferment extract.

Causes of flavor instability of wheat bulgur products are also being studied by gas chromatography. In the early stages of storage deterioration of wheat bulgur shelter wafers, we have detected hydrocarbons in the vapors from the wafers. To augment the study of wheat products, we use model systems, including fatty acid methyl esters. Of the esters treated to promote autoxidation, only methyl linoleate and methyl linolenate produced hydrocarbons. Methyl laurate, methyl oleate, and methyl 11-undecenoate did not produce hydrocarbons over an eight-week period of autoxidation. Thus, it appears that two or more double bonds are necessary in a fatty acid ester for hydrocarbon formation to occur during the initial stages of autoxidation. Oxygen uptake by methyl linoleate suspended in glass wool in an oxygen atmosphere was relatively slow for about a week and then very rapid for about 10 days. It decreased slowly thereafter. Three moles of oxygen gas were consumed per mole of methyl linoleate after 30 days. The high ratio of oxygen consumption strongly suggests that secondary oxidations occur. Gas chromatographic analyses of headspace gases above ground bulgur and ground puffed bulgur stored

under oxygen or nitrogen at four temperatures will be compared with evaluations by odor panels to determine whether any of these oxidized compounds would be useful as advance indicators of rancidification.

7. Pigments of Wheat Bran and Aleurone. Contract research has been initiated at Oregon State University to determine the nature of the substances in bran and aleurone tissue that are responsible for coloration of wheat grain. The inner true bran layers of wheat contain most of the coloring matter of the kernel. Basic studies of bran pigments should be helpful in developing decolorizing procedures for use in whole kernel products. Pigments and phenolic compounds closely associated with pigments have been extracted from bran by sequential leaching with organic solvents and water, and individual compounds separated by paper chromatography after preliminary separations in solvents. Large-scale extractions were made to provide material for further study from genetically pure strains of three varieties of red wheat.

## B. New and Improved Food Products and Processing Technology

1. Bulgur and Related Wheat Products. Recent developments of export markets for bulgur, a parboiled dried wheat used since ancient times in the Near East, have been a major accomplishment in which Department utilization research scientists have cooperated with private industry and grower-sponsored marketing associations. Prior to 1961, there was no significant export market for bulgur. After a trial shipment in 1962 of 60 million pounds for welfare distribution in 27 foreign countries, bulgur exports have grown to supply markets for more than 8 million bushels of wheat per year. Most of this export depends on donations of government surplus and concessional sales, but the product has been well received in many places and efforts are being made to develop a commercial market.

In some markets, particularly in Hong Kong, resistance to the use of bulgur appears to be based on the dark color and high bran content of red wheat bulgur. To produce a quick-cooking, light-colored, bland-flavored product, even from hard red wheat, we have developed a lye peeling method to remove the bran. We call this product WURLD wheat. Pilot-plant operations have been developed to study the process and produce a product for demonstration purposes. About 2500 pounds of WURLD wheat from hard red winter wheat were shipped to Hong Kong for pilot acceptance tests by voluntary welfare agencies there. The preliminary test was gratifying, and requests have been made for more WURLD wheat to evaluate its potential in commercial markets as well as its use for welfare purposes.

WURLD wheat is prepared by precooking wheat, treating it with concentrated alkali to soften the bran adhesion, quenching the alkali in water, scouring the bran by turbulent water flow, separating the peeled wheat from the bran, and neutralizing any residual alkali with acetic acid. After drying, the kernels can be cracked or used whole.

The storage stability of WURLD wheat prepared from white club, hard red spring, and hard red winter varieties was evaluated. After six to eight



months' storage, differences in stability between bulgur and WURLD wheat were small and of no practical significance.

In addition to wheat, barley and other cereals can be peeled down to the aleurone layer by the WURLD process. In this way the loss of material (such as the ends of pearled barley) can be avoided.

A systematic study of the influence of several processing variables on quality of bulgur has been initiated. Soaking time and temperature, steaming time and temperature, and drying conditions are being evaluated as to their effects on the hardness of bulgur, its cooking time, and its expansion by hot-air puffing.

To develop a series of flavorful food products, research has been initiated on the effects of processing variables in the malting of wheat. Temperature and humidity during germination and toasting can be varied to produce a range of flavors and textures that should be useful in developing new food products and richly-flavored flour for blending purposes. In-house research will be supported by contract research recently initiated at the University of Wisconsin on changes in protein and other major nutrients of wheat during malting.

2. Food from Wheat Fractions. In milling wheat into flour, nearly 30% of the kernel becomes byproduct milling fractions, generally used as livestock feed. About two million tons of wheat feed, midlings, shorts, and red dog flour, and about an equal amount of bran are used for this purpose each year in the United States. These mill byproducts contain higher levels of protein than does flour or the starting grain, and the protein is of greater biological value. Research is conducted with the objective of using wheat fractions in new high-protein food products. Substantially all the protein is extractable from finely ground bran at pH 12.5, but the recovery decreases as the pH is lowered. However, only about three-quarters of the nitrogenous compounds can be precipitated from the solutions after extracting; the remainder is non-protein. Treating the bran with cellulose-splitting enzymes or hydrogen peroxide did not improve quality of the protein extracted. Protein recovered from bran by alkali extraction and acid precipitation has a dark brown color, poor stability to rancidification, and a strong branny odor and taste. Modifications in the recovery process will be sought to improve the quality of product.

An improved process was advanced for preparing a bland, milk-like nutritious drink from wheat or wheat fractions high in proteins. After acid-pepsin digestion of flour, prime starch can be separated from other flour components. The material other than prime starch is then heated to bring about gelatinization and partial hydrolysis of the starch fraction it contains. The neutralized and cooled product is a protein-enriched, bland liquid. Viscosity and protein content can be adjusted to those of milk by adding dextrans produced from the prime starch fraction. Drum drying of this milk-like liquid produces a powder that can be easily dispersed in cold water. It is relatively stable and retains most of the nutritional value of the starting wheat protein.



This development of a milk-like product is based, to a considerable extent, on preliminary studies of enzyme digestion of wheat protein conducted under contract at Purdue University. The work at Purdue was concluded after the testing of several procedures to improve the dispersibility and the stability of the dispersion of wheat proteins. Methods examined include deamidation with acid, hydrolysis with protein-splitting enzymes, and chemically induced splitting of disulfide bonds.

3. Emergency Food Supply. Foods suitable for stocking fallout shelters are being developed and evaluated with funds transferred to Agriculture by the Department of Defense. The level of support for this project was substantially reduced from previous years. In-house research includes basic studies on oxidative deterioration and stability of wheat food products (see paragraph 1-A-6). The contract evaluation of stability of bulgur wafers and new food adjuncts for a wafer-based ration continues at Oregon State University. Taste panel evaluations of bulgur wafers after 22 months of storage and chemical analyses after 16 months stored at three temperatures indicate that product stability at this stage is satisfactory even up to 100° F. Packing in nitrogen is preferable to packaging in air, and formulation with malt syrup is preferred to formulation with corn syrup. Differences in chemical tests do not, at this stage, show trends consistent enough to correlate with test panel evaluations. Such correlation is sought for use in developing a stockpile surveillance procedure.

4. Baking Quality of Flours. To provide improved testing procedures for baking quality of flours, to assist in development of improved varieties, and to check the accuracy of our test procedures, we are collaborating with the Hard Winter Wheat Quality Council in comparing values obtained from farinograms, mixograms, and pressure-meter measurements of diastatic activity from straight doughs made by a 3-hour fermentation procedure. Included in the tests this year were samples of 26 hard winter wheat flours from six states. A major function of the Council is to recommend release of new wheat varieties for commercial production. An important benefit to us resulting from this collaboration is the evaluation of our test procedures. Since our results have been comparable with those of most other testers, we believe we can use our methods with confidence to evaluate bread-baking performance of high-protein fractions obtained by air classification of hard red winter wheat flours and blends of high-protein fractions with low-protein flours.

We are currently testing low-protein fractions from air-classified flours for performance in cakes. Low-protein fractions from five different wheat flours were bleached with chlorine gas. The treatment produced a different pH in each flour. The cause of these differences in chlorine-induced pH changes has not yet been learned, but the pH change does not correlate with the relative surface area of the flour, the proportion of gluten in it, the proportion of low-buffered prime starch, or the ash content--four factors that have been mentioned by others as having an effect on the chlorine requirement for bleaching flour to a specific pH.

Continuous-mix bread-making processes, in general, tolerate less nonfat milk in formulation than do sponge and dough methods, and some flours affect this tolerance more than do others. Even though flour and milk present two very complex protein systems, we are making headway in that we can see an implication of an active protease in flour capable of splitting a specific casein fraction of milk. The amount and activity of the flour protein enzymes, which vary from one flour to another, may be responsible for the variations in milk-carrying capacity of a continuous mix dough. We will isolate the major protein components from both the flour and the nonfat dry milk to provide sufficient quantities to incorporate in doughs for tests to provide evidence of specific effects produced by different protein components. For continuing studies of the effects of milk protein on continuous-mix bread, mixing and baking procedures are being standardized by use of the Brabender Do-Corder.

The effect upon baking quality of variation in wheat flour lipids was investigated under a P.L. 480 grant to the British Baking Industries Research Association in Chorleywood, England. They studied the influence of variety, season, and environment on the composition and baking quality of flours from five wheats grown in the United States and two wheats grown in Great Britain in the crop years of 1959-1962. The varieties did not differ with respect to the number and types of lipids present, but the quantity of these lipids varied substantially with variety, season, region, and environment. Apparent correlations between analytical data and baking performance were not consistent from one season to the next. Baking tests demonstrated that fat in the presence of the optimum amount of an oxidizing agent can exert an improving effect on bread, and a pure synthetic saturated lipid had a similar effect.

Another P.L. 480 grant has been awarded to the same institution to study the structure of dough and baked products as a function of interactions between dough constituents, especially the interaction of lipids. This new investigation will build on the foundation of competent research developed in the first grant.

5. Carrying Capacity of Hard Red Winter Wheat. In Western European countries large amounts of wheat are grown that are too soft and of too low a protein content to make high-quality yeast-raised bread. To add strength and tolerance to doughs and give uniform baking qualities, these countries import wheat to mix with their locally-grown wheat, but some of them restrict the amount of imported wheat that may be used. The hard wheats differ in their carrying capacities, that is, the degree to which a given percentage of hard red wheat will improve the soft wheat. Flours from the flinty, vitreous hard wheats are granular and sharp compared with flours from soft wheat, and inadvertent classification may occur during or after blending. Furthermore, different types of hard wheats react differently when blended with soft wheat and mixed into dough for bread baking. In order to provide information that may expand markets for surplus United States hard winter wheats, we are investigating the carrying capacity of the hard U.S. winter wheats. Research on blending characteristics was initiated by contract at Kansas State University with



samples of soft wheats procured from Germany, England, Sweden, The Netherlands, Austria, France, and Belgium and samples of hard red winter and spring wheats from the United States and Canada. Flours milled from these wheats will be blended to provide a range in protein content and evaluated for baking and dough-mixing properties.

6. Nutritive Value of Processed Wheat. In developing wheat food products, attention is given to the highest possible degree of nutrient retention. In WURLD wheat, which was developed to overcome objections to the color and fiber content of bulgur, the bran and germ normally present in bulgur are removed by an alkaline treatment that reduces the crude fiber content and lightens the color of the product. The process is designed to retain as much of the aleurone tissue as possible because of its high nutritive value. Average levels of thiamine, riboflavin, vitamin B<sub>6</sub> and folic acid in WURLD wheat are somewhat lower than in bulgur made from the same wheat, but more than half of the amount originally contained in the wheat is retained. Even greater proportions of niacin, choline, and pantothenic acid are retained. Protein efficiency ratios for WURLD wheat are only slightly less than for bulgur. Although these values represent lower levels of nutrients than in whole ground wheat, they exceed by far those in white flour.

Rapid chemical methods for assay of the biological value of wheat proteins during processing are being developed under P.L. 480 at Cambridge University in England. A chemical method or a combination of chemical methods that will correlate with rat- and chick-feeding tests for availability of protein in wheat foods is sought. The success of this work would simplify and reduce the cost of biological evaluations and should provide, also, a substantial increase in the amount of data available for use in improving food products. A chemical analysis for "total tryptophane" has been partially successful as an indication of protein efficiency ratio for some products, but not for wheat flour or wheat products. Several attempts to reduce hydrolysis losses in determining available lysine were made, but none of the approaches used was completely successful. As a result, chick-feeding tests are still being used to determine effects of processing treatments on nutritional quality of wheat products.

When wheat is steeped in hot water, its protein efficiency ratio appears to be increased but the effect later proved to be due to more efficient use of carbohydrate present. Such steeping is a preliminary step in both bulgur and WURLD wheat production.

7. Microbiology of Wheat Food Products. Relative freedom from microbial contamination of flour to be used in precooked frozen foods, baby foods, and certain canned products is an important safety factor for such foods. Under contract research, the American Institute of Baking in Chicago is determining the nature and extent of microbial contamination in wheat flour and studying means to reduce or eliminate it. Flour inoculated with four types of contaminating micro-organisms at the level of one million organisms per gram



has been treated with propylene oxide. The treatment destroyed the most resistant of the micro-organisms, B. subtilis, even in its stabilized spore form. No significant damage to flour properties was found, but the economics of the treatment are yet to be worked out.

Survival patterns of flour-introduced microbial contaminants were determined in frozen soups and pie doughs. E. coli and A. flavus contaminants were destroyed in the food processing operation, or they survived for less than a week in frozen storage. S. aureus and B. subtilis were virtually unchanged as a result of the freezing and storage treatments.

#### C. New and Improved Feeds and Feed Processing Technology

1. Improved Feeds from Wheat and Wheat Fractions. Wheat milling byproducts contain important nutrients at levels of concentration higher than in milled flour, but because of general variability in quality and condition, they cannot compete with protein, carbohydrate, and other nutrients from other sources for use in mixed feeds. The unrealized feed values in mill byproducts adversely affect milling profits and are reflected in higher flour prices as well as in pressure to reduce wheat prices to growers. Research is being conducted on enzymic release of carbohydrates from cellulosic components of mill run. Preliminary results of using salivary amylase were promising. Bacterial and fungal amylases did not work well. Research is underway to provide better analytical methods for estimating biological value of mill run fractions.

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FORAGES AND FEED - PROCESSING AND PRODUCTS - NORTHERN REGION  
Northern Utilization Research and Development Division, ARS

Problem. The demand for livestock in the United States will increase 45 percent by 1975. There is an increasing demand for processed forages in European and Asiatic markets. Fresh forage crops are the richest natural source of many nutrients for farm animals. The bulk of forages, however, is preserved so inefficiently by haymaking and ensiling that 10 to 50 percent of the dry weight, and much larger fractions of the most valuable nutrients, are lost before the animals eat them. Dehydration is now the only practical means of preserving the high nutritional value. Poultry and swine producers are aware of the value of dehydrated forage, but restrict their consumption because of high fiber and growth-inhibitor content. The livestock breeder needs forage products tailored to specific animals, and the forage producer must adapt to his needs to sell.

Basic and applied utilization research are necessary to produce: (1) nutritional juice and low-fiber, high-protein feed for non-ruminant animals; (2) fiber products which have been cheaply treated to make them easily digestible for ruminants; (3) growth-stimulating supplements for ruminants based on the biologically active fiber-digestion factor, and growth-promoting factor in forage. Further, new products should be adaptable to mechanical feeding. Improved handling will encourage farmers to put high-value land now producing surplus crops into forages.

USDA AND COOPERATIVE PROGRAMS

The major part of the Department's research program on forages is maintained at the Western Utilization Research and Development Division, Albany, California. At the Northern Division, Peoria, Illinois, the Department has a short-term program involving one organic chemist engaged in research to isolate and identify the toxic component(s) of tall fescue grass responsible for a cattle disease known as "fescue foot." This research is cooperative with the Kentucky State Experiment Station which furnishes toxic and nontoxic fescue grass for chemical study and conducts bioassays of fractions and components isolated from fescue at the Northern Division. Liaison is maintained with the fescue breeding program of the Field Crops Research Branch, ARS, through the Agronomy Department of the University of Kentucky and with the Department's Pharmacology Laboratory at the Western Division.

The Federal program at Peoria, Illinois, totals 1.3 professional man-years, all of which is devoted to chemical composition and physical properties.

## PROGRAM OF STATE EXPERIMENT STATIONS

State stations conduct an extensive program of both basic and applied research on forage utilization. Much of the research is interdisciplinary and often involves several departments.

One major segment of the research effort is devoted to determining the chemical composition of forages and evaluation of the relationship between chemical composition of certain forages and their nutritive value for farm animals. Evaluation of the effects of certain agronomic, cultural, processing and handling practices on composition, palatability and nutritive value of forages receives much research attention. Fiber content and utilization of fiber by swine, cattle, sheep and poultry affect the value and use of forages. Methods of isolating and analyzing for fiber are being developed. Investigation of normal and abnormal rumen fermentations of forages is fundamental to maximum utilization.

Careful studies of specific constituents of forages are being undertaken. Determination of certain minor elements found in forages is important both from nutrition and toxicity standpoints. Protein content and quality merit special attention along with determination of amino acid values and unknown growth factors. Leaf organic acids and proteins are investigated in detail in an effort to increase our understanding of their biosynthesis and properties in relation to growth of forage plants.

Due to the economic importance of forages in animal feeds, development of means for evaluation of the nutritive quality of forages has become an important field of study. New and more accurate or rapid chemical procedures are being sought.

Development of forage handling and processing systems to minimize labor costs has led to increased research on forage processing methods. Fermentation characteristics of and animal response to forages which have been wilted, chopped, pelleted, ensiled or dehydrated are being determined. Small-scale ensiling systems are being used to evaluate various silage preservatives. Methods of dehydrating alfalfa are being studied and the economic feasibility of dehydration is being investigated.

The total research effort devoted to forage utilization is about 22.7 professional man-years.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Chemical Composition and Physical Properties

1. Fescue toxicity. Work on this project is now completely reoriented to test the hypothesis that mycotoxins may be the cause of fescue foot in cattle. The Northern and Western Divisions and Kentucky AES are cooperating

in this study and maintaining close contact for planning and exchange of information.

Survey of toxic fescue hays for types of fungi present has been initiated. Some 35 strains of fungi from fescue hays have been identified. Fescue hay inoculated with selected mold organisms and then incubated, and extracts of several fungi and of toxic hay have been sent to the Western Division for use in developing and evaluating small animal bioassay methods.

#### PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

##### Chemical Composition and Physical Properties

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FORAGES AND FEED - PROCESSING AND PRODUCTS - WESTERN REGION  
Western Utilization Research and Development Division, ARS

Problem. The demand for livestock in the United States will increase 45% by 1975. Forage crops constitute the major feedstuff for ruminant animals. In addition, there is an increasing demand for processed forages in European and Asiatic export markets. Fresh forage crops are the richest natural source of many nutrients for farm animals. Forages, however, are preserved so inefficiently by haymaking and ensiling that 10 to 50% of the dry weight, and much larger fractions of the most valuable nutrients, are lost before the animals eat them. Dehydration is now the only practical means of producing high nutritional value products in a form usable in manufactured feeds and supplements. Poultry and swine producers are aware of the value of dehydrated forage, but restrict their consumption because of high fiber and growth-inhibitor content. There is evidence to show that certain unidentified growth factors are at least partially lost during the dehydration process as presently carried out. The livestock breeder needs forage products tailored to specific animals, and the forage producer must adapt to his needs to sell.

Basic and applied utilization research are necessary to produce: (1) high protein, low-fiber feeds rich in unidentified growth factors designed for use by non-ruminant animals; (2) fiber products which have been cheaply treated to make them easily digestible for ruminants; (3) growth stimulating supplements for ruminants based on the biologically active fiber digestion factors and growth-promoting factors in forage. New products should be adaptable to mechanical feeding. Improved uses will encourage farmers to put high-value land now producing surplus crops into forages.

USDA AND COOPERATIVE PROGRAM

Current research in the Western Utilization Research and Development Division includes both basic and applied studies on all forages used or potentially usable for off-the-farm processing. The research is conducted at the Division headquarters at Albany, California; under contract at Berkeley, California; Lincoln, Nebraska and Athens, Georgia; and under the P.L. 480 grant programs in Scotland and Italy. Basic compositional studies deal with the potent estrogen, coumestrol (discovered by Department scientists), and other phenolic compounds present in forage legumes. The value of coumestrol-rich alfalfa as a growth stimulant for ruminants is being studied with financial support of the Department of Agriculture and Inspection of the State of Nebraska and the cooperation of ARS Farm Research and several experiment stations and commercial processors of forages. Also under study are other biologically active forage constituents (such as the chick-growth-promoting factor in forage juices and alfalfa saponins which depress chick growth), organic acids of alfalfa, and the mechanism of action of forage

antioxidants. Processing of forages by "wet" (juicing) and "dry" (turbo-milling and air classification) methods is being investigated.

The Federal program of research in this area totals 9.9 professional man-years, including one scientist whose salary is provided by the Department of Agriculture and Inspection, State of Nebraska, and contract research equivalent to 2.8 professional man-years per year. Of this number 4.7 are assigned to chemical composition and physical properties; and 5.2 to new and improved feeds and processing technology. In addition the Division sponsors, under P.L. 480, two research projects on forage composition.

#### PROGRAM OF STATE EXPERIMENT STATIONS

State stations conduct an extensive program of both basic and applied research on forage utilization. Much of the research is interdisciplinary and often involves several departments.

One major segment of the research effort is devoted to determining the chemical composition of forages and evaluation of the relationship between chemical composition of certain forages and their nutritive value for farm animals. Evaluation of the effects of certain agronomic, cultural, processing and handling practices on composition, palatability and nutritive value of forages receives much research attention. Fiber content and utilization of fiber by swine, cattle, sheep and poultry affect the value and use of forages. Methods of isolating and analyzing for fiber are being developed. Investigation of normal and abnormal rumen fermentation of forages is fundamental to maximum utilization.

Careful studies of specific constituents of forages are being undertaken. Determination of certain minor elements found in forages is important both from nutrition and toxicity standpoints. Protein content and quality merit special attention along with determination of amino acid values and unknown growth factors. Leaf organic acids and proteins are investigated in detail in an effort to increase our understanding of their biosynthesis and properties in relation to growth of forage plants.

Due to the economic importance of forages in animal feeds, development of means for evaluation of the nutritive quality of forages has become an important field of study. New and more accurate or rapid chemical procedures are being sought.

Development of forage handling and processing systems to minimize labor costs has led to increased research on forage processing methods. Fermentation characteristics of and animal response to forages which have been wilted, chopped, pelleted, ensiled or dehydrated are being determined. Small-scale ensiling systems are being used to evaluate various silage preservatives. Methods of dehydrating alfalfa are being studied and the economic feasibility of dehydration is being investigated.

The total research effort devoted to forage utilization is about 22.7 professional man-years.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Chemical Composition and Physical Properties

1. Chemical Constituents of Forages. Research is conducted on phenolic components of alfalfa and clovers. Twelve phenolic compounds already have been isolated and fully characterized. Coumestrol was the first and was reported in previous years; the other compounds have structures similar to coumestrol. Biological activity of compounds of this type include effects like those of female hormones. Coumestrol was initially isolated by following estrogenic activity of alfalfa fractions (see paragraph 3-B-1). The estrogenic and other biological activity of these compounds will be under continuing study.

2. Interactions of Forage Antioxidants. Carotenes, vitamin E, xanthophylls, and other related substances, provide much of the biological value of alfalfa. These substances gradually deteriorate when exposed to oxygen. We are trying to understand natural antioxidant activity in alfalfa to guide us in developing products that will not deteriorate. Contract research is conducted at the University of California in Berkeley on separation of alfalfa lipids and the relationship of lipids to carotene oxidation in forage crops. Mixtures of galactolipids were isolated by thin-layer chromatography. Galactolipids made up half the total alfalfa lipids, and methodology for separating mono- from di-galactolipids was improved. Two-dimensional thin-layer chromatography separated 18 components, but further improvements are required to separate and quantitatively recover individual lipid components. The extreme instability of these compounds complicates their separation. Light-induced changes and oxidation may alter these lipids before they are separated.

Under a P.L. 480 grant, research at the Experiment Station for Practical Agriculture in Milano, Italy, is underway on non-tocopherol reducing substances in alfalfa to throw light on the natural antioxidant activity of alfalfa. To prevent oxidative changes during separations in laboratory research, synthetic antioxidants are used as oxygen acceptors. They protect, for instance, carotenoid double bonds from peroxidation. Alpha-tocopherol has natural antioxidant activity. Paper chromatography separated and detected alpha-tocopherol and other antioxidant substances. One of the other reducing substances is plastoquinone. Surveys of reducing substances were made with alfalfa at different stages of maturity. Alpha-tocopherol and other substances under investigation tend to increase in amount as the plant matures. The distribution of reducing compounds in various parts of the plant was also surveyed. The stem had the same general composition as the leaves, but with reducing substances in far lower concentration. In yellow leaves, one specific compound was particularly prevalent, but this compound was absent in the flowers; it exists only in the chloroplasts.



3. Structure of Alfalfa Polysaccharides. Apparently combination with indigestible lignin reduces the digestibility of alfalfa polysaccharides. Research is being conducted under P.L. 480 funds at Edinburgh University in Scotland to determine the nature of alfalfa polysaccharides and investigate enzyme systems that may be helpful in structural analysis of alfalfa polysaccharides and components associated with them. Enzymes, which were extracted from alfalfa seedlings, could hydrolyze still other carbohydrates. Polysaccharide fractions were isolated from separated leaves and stems and the main structural linkages were determined for the stem hemicellulose.

B. New and Improved Feeds and Processing Technologies

1. Coumestrol-Enriched Feeds. Tests on growth response of sheep to pure crystalline coumestrol were completed at the Oregon Experiment Station in cooperative studies. They confirmed the growth-promoting effect of coumestrol which had been previously found by feeding alfalfa meal rich in coumestrol. Lambs on a ration containing coumestrol used feed 11% more efficiently and gained weight 18% faster than did lambs on the basal ration. The Indiana Experiment Station and the Animal Husbandry Research Division of the Agricultural Research Service are cooperating in the evaluation of coumestrol as a growth promoter in large animal feeding trials, but no conclusions have been reached yet.

2. Improved Alfalfa Meal. Studies on the improvement of alfalfa products by wet and dry fractionation procedures are supported in part by the Nebraska Department of Agriculture which provides the salary of one scientist. In cooperation with the Dixon Dryer Company, Dixon, California, experimental equipment for combined water extraction and dehydration of forages was installed at their plant. Successful runs provide data for planning expanded studies during the 1965 crop season. Some of these experiments on dry processing will be conducted in Nebraska. A supply of concentrated water extract from alfalfa was obtained for chemical characterization and bioassay. A separation milling procedure for alfalfa has been developed and was tested in 1964 on 20 cuttings of three plots of alfalfa harvested at intervals of 26, 30, and 34 days. Each sample of each cutting was processed to separate stem from leaf and analyzed for proximate feed composition, carotenes, and xanthophylls. By separating leaf meal, plants can be allowed to mature further and yield higher tonnage per acre without sacrifice of feed quality. Large samples were retained for bioassay of xanthophyll availability. A preliminary experiment indicated that the amount of pigmentation produced in poultry is markedly improved by pelleting and regrinding alfalfa leaf fractions. Large amounts of two types of saponins were prepared from alfalfa for further bioassay. Dimethylsulfoxide was useful to dissolve difficult saponins for bioassay with chicks.

3. Products from Southeastern Grasses. Contract research was initiated at the University of Georgia to study two southeastern forage crops, coastal Bermuda grass and Pearl millet. Four cuttings of Bermuda grass were made to

obtain forage of different maturity levels and six cuttings of millet. From each, samples were prepared of fresh chopped forage, dehydrated chopped forage, and pelleted meal with different antioxidant and antioxidant synergist treatments for use in stability studies. Carotene, xanthophyll, and tocopherol composition will be studied throughout storage tests. For the 1964 growing season in the area tested, millet had higher protein and higher carotene concentration than coastal Bermuda grass. Seasonal changes may be important so several years' testing are required for conclusive evidence.

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Chemical Composition and Physical Properties

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RICE - PROCESSING AND PRODUCTS - SOUTHERN REGION  
Southern Utilization Research and Development Division, ARS

Problem. The productive capacity of U. S. rice growers has increased faster than domestic and export consumption over the past decade, thus limiting the income potentially available from this major world food grain. Detailed knowledge of chemical composition and physical properties, as related to processing is needed to guide milling, processing and product development of U. S. rices so that they can better meet the quality and new product requirements needed for expanded markets. New and diverse food products from rice that are easy to prepare, have flavor and texture appeal, and are economical to manufacture, are needed to increase the total consumption of rice both domestically and abroad. Additional needs include the development of improved milling machinery and techniques, primarily to increase the yield of head rice; intensified research on deep milling to evaluate and utilize the products, protein flour and residual kernels; and research to provide greater flexibility in the industry by developing from either medium or long grain rice new products that will provide on cooking either discrete kernels or a gelatinous food.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program involving at New Orleans, Louisiana, biochemists and analytical chemists engaged in basic and exploratory studies on the distribution of the chemical constituents of milled rice in consecutive layers of the kernel with special emphasis on nutritionally important constituents such as proteins, amino acids, starch, lipids, vitamins and minerals; and on the cooking and chemical characteristics of the kernels remaining after differential removal of these layers. Findings from this research will provide the necessary basis for evaluating the economic feasibility of using high-protein rice flours (layers removed by deep milling) for protein fortification of foods and as dietetic or other specialty type foods.

Close cooperation is maintained, under formal memoranda of understanding, with the Louisiana, Arkansas and Texas Rice Experiment Stations, who supply rice samples of known variety and cultural history for the experimental studies. The Rice Inspection Service, Grain Division, AMS, New Orleans, Louisiana, cooperates by providing assistance in grading rice samples from the research investigations. Cooperation has been initiated with the Western Division.

The Federal scientific effort at the Southern Division devoted to research in this area totals 2.7 professional man-years. The present effort is on chemical composition and physical properties.

## PROGRAM OF STATE EXPERIMENT STATIONS

Rice research in progress at the State stations involves evaluation of new rice selections and varieties for agronomic and milling quality. Emphasis is placed upon developing and applying rapid, simple testing procedures useful for screening selections. Much of this work is carried out in cooperation with the Regional Rice Quality Laboratory. Laboratory methods for determining the quality of milled rice are sought for use in establishing objective standards for measurement of the quality of cooked rice. Cultural practices that may affect the quality, drying properties and storage stability of rice are evaluated to determine their influence upon processing characteristics and product quality.

Basic composition studies relate to the quantity and quality of the proteins, lipids and starch fractions and to their distribution within the kernels. Occurrence of mycotoxins in rice is being studied. Attempts to develop methods for the prevention and control of mycotoxin elaboration in rice during processing and marketing continue.

Product research involves development of effective ways to use rice in quantity food service. Another study seeks ways to make more efficient use of rice proteins through study of the supplementary value of high protein foods derived from rice and its by-products. The biological value of the proteins of rice, when used with multipurpose food is being investigated. This research is especially timely in view of the potential for use of rice in the diets of developing countries.

The influence of drying methods used on rough rice on the processing characteristics of rice is under study. Other variables such as maturity and variety are also studied.

The total State scientific effort devoted to utilization of rice is 1.9 professional man-years.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Chemical Composition and Physical Properties

1. Investigation of the Distribution of Chemical Constituents in the Rice Kernel. In continued study of the distribution of the chemical constituents of the rice kernel, 12 consecutive peripheral layers of commercially milled Bluebonnet rice, totaling 19% of the kernel's weight, were removed by deep-milling in a prototype mill employing the tangential-abrasion principle. The protein, riboflavin, thiamine, niacin, phosphorus, and calcium contents of the flour decreased in the respective layers from the periphery toward the center. The level of each of these constituents in the outer layers greatly exceeded the level in the original kernel. The protein decreased from 22 to 12% in the 12 layers from periphery to center, but the amino acid pattern of the protein in the respective layers did not change, except for tryptophan,



which decreased by about 40% from layer 1 to layer 12. Although the total amount of protein present in the original and residual kernels differed by about 3%, the amino acid patterns of the proteins were identical. In comparison with wheat and corn proteins, these rice proteins contain the same or higher percentages of amino acids essential for nitrogen balance in adults, except for leucine, which is highest in corn.

The protein-rich rice flour holds promise in baby, geriatric, nonallergenic, and other dietary foods and in entirely new rice products. The residual kernels should be of special value for the manufacture of beer, wine, starch, and cosmetic preparations. Since wheat (hard winter and soft red), barley, and grain sorghum milled on a laboratory device showed high protein peripheral layers in all except soft red wheat, it appears that several products having greater nutritive value than the original grains may be prepared for human consumption, should deepmilling prove commercially feasible. (Sl 4-13).

#### PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

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RICE--PROCESSING AND PRODUCTS--WESTERN REGION  
Western Utilization Research and Development Division, ARS

Problem. The productive capacity of U.S. rice growers has increased faster than domestic and export consumption in recent years to cause imposition of acreage limitations and restrictions in the income potential for growers and the U.S. economy. New and diverse food products from rice that are easy to prepare, have greater texture and flavor appeal, and are economical to manufacture are needed to increase the total consumption of rice both here and abroad. Likewise needed are drastically improved milling methods to increase economic returns to growers and millers from the limited production allowed. Detailed knowledge of chemical composition and physical properties, as related to processing, is needed to guide the developments in milling, processing, and products required to achieve expanded markets.

USDA AND COOPERATIVE PROGRAM

In the Western Utilization Research and Development Division, basic and applied research on rice is conducted at Albany, California. Basic studies involve chemical, physical, and biochemical investigations of rice proteins and carbohydrates and changes undergone by these constituents during processing. Process development is underway on debranning of brown rice by lye-peeling, parboiling of brown and undermilled rice, new methods to produce quick-cooking forms having better flavor and texture, and on conversion of high-protein flours into beverage products especially suitable for infant feeding overseas. New gloss-producing agents to replace talc for coated rice are in final stages of commercial-scale testing.

The Federal program of research in this area totals 2.9 professional man-years. Of this number 1.1 are assigned to chemical composition and physical properties; 1.8 to new and improved food products and processing technology.

PROGRAM OF STATE EXPERIMENT STATIONS

Rice research in progress at the State stations involves evaluation of new rice selections and varieties for agronomic and milling quality. Emphasis is placed upon developing and applying rapid, simple testing procedures useful for screening selections. Much of this work is carried out in cooperation with the Regional Rice Quality Laboratory. Laboratory methods for determining the quality of milled rice are sought for use in establishing objective standards for measurement of the quality of cooked rice. Cultural practices that may affect the quality, drying properties and storage stability of rice are evaluated to determine their influence upon processing characteristics and product quality.

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Occurrence of mycotoxins in rice is being studied. Attempts to develop methods for the prevention and control of mycotoxin elaboration in rice during processing and marketing continue.

Product research involves development of effective ways to use rice in quantity food service. Another study seeks ways to make more efficient use of rice proteins through study of the supplementary value of high-protein foods derived from rice and its by-products. The biological value of the proteins of rice, when used with multipurpose food is being investigated. This research is especially timely in view of the potential for use of rice in the diets of developing countries.

The influence of drying methods used on rough rice on the processing characteristics of rice is under study. Other variables such as maturity and variety are also studied.

The total State scientific effort devoted to utilization of rice is 1.9 professional man years.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Chemical Composition and Physical Properties

1. Rice Proteins. Rice protein has a high biological value relative to other cereal proteins. We are investigating the amino acid composition of two globulin fractions. One fraction precipitates at pH 4, contains two components, and has almost no lysine or histidine. The other, soluble at pH 7, has five or six components and is rich in sulfur-containing amino acids. We are separating components of these fractions for further study.

About 0.4 to 0.7% of rice protein has been extracted as prolamine, which is distinctly different from the globulins. It contained some 20% of non-protein material. The entire prolamine fraction has been divided into 7 components by electrophoresis.

##### B. New and Improved Food Products and Processing Technology

1. Improved Rice Products. Deep milling of white rice to produce a flour having double the protein content of the starting, normally-milled kernel was reported a year ago. Development of this high-protein rice flour is being investigated at both the Southern and the Western Utilization Research Divisions. At the Western Division we are comparing amounts and types of proteins in the flour with those in the original rice and working on procedures using commercially available equipment for deep milling to increase the protein content of the rice flour. A single-pass abrasive milling with equipment similar to certain types available commercially, yielded 3-4% of high-protein flour. Breakage was 2% or less. Second heads (large broken kernels) provided somewhat less than a 2-fold protein increase. Air



classification of defatted rice polish and bran provided some concentration of protein in the product fractions.

Rice flour containing 15-1/2% protein has been shipped to the United Nations Children's Fund for protein evaluation by animal feeding tests and for accompanying analyses. Because rice is the common man's food in many areas of the world where protein deficiency is a major nutritional problem for young children, the deep milling of rice to provide a protein-rich food is potentially a very significant development of utilization research.

A high-gloss rice is preferred in the Caribbean markets where much California-grown rice is sold. High gloss is customarily achieved by coating the milled rice with glucose and talc, but since talc is not metabolized, regulatory agencies are concerned about its use. We have been testing a number of alternative materials for use in the sheen-coating. Limited commercial trials with calcium citrate or calcium acetate in place of the talc appear promising. The acetate gave less sheen than does talc, but the citrate produced a sheen nearly like that from talc. Taste panels were unable to detect any flavor difference related to the citrate coating.

Color formation during processing of parboiled rice is generally believed to be due to the browning reaction, but pigment from the hull and bran may also contribute substantially to the overall color. In an investigation of the effects of processing variables in parboiling brown or undermilled rice, we found that increased pressure during cooking yielded parboiled rice with a lower percentage of broken kernels, but color of the rice was darker when higher cooking pressures or longer cooking times were used.

Brown rice can be peeled down to the aleurone layer by the WURLD process. Such rice is absolutely white. Even in a raw form it can be steamed in 20 minutes for eating. It should have a higher retention of the protein, vitamins, and other nutrients known to be located in the aleurone layer.

PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

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## NUTRITION AND CONSUMER USE RESEARCH

Consumer and Food Economics Research Division, ARS  
Human Nutrition Research Division, ARS

Problem. The assortment and characteristics of foods available to consumers change constantly with the adoption of new practices of production, processing, and marketing. Changing constantly also, as nutrition science advances, is our understanding of the nutritional needs of man and the manner in which these needs can best be met by food. To help meet the Department's responsibility to advise consumers on the quantity and variety of foods that will assure maximum benefit and satisfaction research must continue on the nutritional requirements of persons of all age groups, on the nutrient and other values of foods and on how to conserve or enhance these values in household and institutional preparation and processing. Periodic surveys of the kinds, amounts, and costs of foods consumed by different population groups and individuals also are essential to evaluate the nutritional adequacy of diets and to give the guidance needed for effective programs of nutrition education. Information from such surveys provides assistance needed in market analyses for different commodities and in the development and evaluation of agricultural policies relating to food production, distribution, and consumer use.

## USDA AND COOPERATIVE PROGRAM

The Department has a continuing program of research concerned with (1) nutritive and other consumer values of raw and processed foods as measured by chemical or physical means and by biologic response; (2) effects of household practices upon the nutritive values and inherent qualities of foods, and the development of principles and improved procedures for household food preparation, care, and preservation; (3) surveys of kinds, amounts, and costs of foods consumed by different population groups and the nutritional appraisal of diets and food supplies; and (4) development of guidance materials for nutrition programs.

The research is carried out by two divisions of the Agricultural Research Service -- the Human Nutrition and the Consumer and Food Economics Research Divisions. Most of the work is done at Beltsville and Hyattsville, Maryland; some is done under cooperative or contract arrangements with State Experiment Stations, universities, medical schools, and industry. The total Federal scientific effort devoted to research in these areas is 77.5 man-years. It is estimated that 14.8 man-years is concerned with studies related to grain products.



Human metabolic studies and the related exploratory and confirmatory studies with experimental animals and microorganisms concerned with defining human requirements for nutrients and foods are not reported on a commodity basis, though some of the work is applicable to this report. This basic nutrition research represents a total Federal effort of 30.2 professional man-years and is described in detail in the report of the Human Nutrition Research Division. Certain aspects of this research related to wheat are considered in this report.

#### PROGRAM OF STATE AGRICULTURAL EXPERIMENT STATIONS

Nutrient value of foods. Research on the nutritive composition of food is often concentrated on locally produced commodities. With the increase of processed and prepared foods, much of the evaluation is related not only to genetic factors and feeding practices but to changes induced by processing and storage to arrive at a value which represents the dietary contribution of the product. Determination of protein and amino acid content and vitamin content of food are active research areas.

The total program in this area includes 38 projects in 24 States and is comprised of approximately 27.3 professional man-years.

Properties related to quality and consumer use of food. Research on food preparation for consumer use may be accompanied by measures of quality from the raw state, through handling and processing for marketing, to final home and institutional service. Special measures characterize certain classes of products; e.g., vitamin assays, enzymatic activity, water binding capacity, and changes in structural tissues.

The structure of baked products as related to the physical and chemical properties of the starches used and supplementary products involved as fats and sugars are the subjects of ongoing basic research in the carbohydrate area. The physical structure of frozen and stored batters and doughs is under study.

Special research emphasis is on the physical and chemical alterations involved in home preparation of foods. This work is carried out with the objective of having foods of maximum quality and nutritional value for final consumption. These researches include suitable methods for home freezing and storage of fresh and precooked foods; special conditions involved in high altitude cooking and baking; the effect of the use of saturated and unsaturated fats and oils on the quality of the final product baked at variable altitudes; and flavor characterization in frozen and stored products by means of vapor component identification. Many of these same factors are being investigated for institutional preparation where the quantities involved impose special conditions.

The State program in this area includes 55 projects in 27 States and involves approximately 50.5 professional man-years. This is a partial report of the State Experiment Station program in food science and includes the work undertaken and participated in by Departments of Home Economics. For research on food and fiber, see the reports of the Utilization Research and Development Divisions, and Clothing and Housing Division.

Food consumption and diet appraisal. The State program in food consumption and diet appraisal extends the work of the Department to selected segments of the population or to smaller geographic areas. One continuing investigation in the North Central Region is planned to yield information regarding food purchase and consumption patterns of families with preschool children. This research will provide information of use to both consumer education and market interests.

A continuing consumer panel in a Southern metropolitan area is providing information on purchase patterns, including data on changes in form, amount, kind, expenditure, and nutritive value of foods purchased. Attempt is made to identify and quantify in a relative sense the factors effecting change.

Currently 16 States are contributing to the Experiment Station program in this area which totals 22.7 professional man-years.

#### PROGRESS -- USDA AND COOPERATIVE PROGRAMS

##### A. Nutrient and Other Consumer Qualities of Wheat and Wheat Products

Two new projects are focused on nutrients in wheat products. Research on the content of nutrients in a wide variety of wheats and wheat products has been initiated, extramurally under research contracts with the American Institute of Baking at Chicago, Illinois, and The Purdue Research Foundation at Lafayette, Indiana, and intramurally at Beltsville. Different varieties of wheat from the major regions of the United States where they are grown will be analyzed before and after regular and air classification milling procedures. Their baked products, which include breads by intermittent and continuous dough procedures, will also be analyzed. The study is planned to show the effects of processing on nutrients of wheat products, including study of the newer milling and baking procedures. Identical samples will be analyzed for many nutrients including thiamine, riboflavin, free and bound niacin, vitamin B<sub>6</sub> components, the individual amino acids, fatty acids, tocopherols, a number of mineral elements, and the proximate components. In addition, a wide variety of wheat products available to the consumer in 10 locations throughout the United States will be analyzed similarly. Products will include bread, cakes, doughnuts, flour, and breakfast cereals.

Preliminary studies have been made on the extraction of lipid materials from wheats and wheat flour prior to fatty acid or tocopherol analysis. Incomplete recovery of the lipids and much of the variability in the data may be minimized with improved extraction procedures. The lipid studies also included development of procedures for determining the individual tocopherols using gas-liquid chromatography. Determination of the individual forms is important because whole wheat contains several tocopherols of varying biological activity as well as an important amount of  $\alpha$ -tocopherol.

Installation of recently acquired atomic absorption equipment should enhance the determination of trace mineral elements in the wheat studies. The new equipment shows promise for yielding more reliable data on mineral elements in foods than those given by the arc spectrograph.

Under the pesticide program, studies have been initiated on possible implications for nutritional and other consumer qualities of repeated use of fumigants on stored wheat. Methyl bromide, ethylene dichloride-carbon tetrachloride (3:1), and phosphine are the fumigants. The wheat will be stored for 3 years, with fumigation every 6 months. Unfumigated control samples stored at ambient and at refrigerated temperatures parallel the fumigated samples. Nutrient analyses will be made for thiamine, riboflavin, niacin, vitamin B<sub>6</sub> components, individual tocopherols, and proximate components in the wheat, the flour, and milling fractions, as well as bread and rolls baked in the laboratory. Bread and rolls and doughs from these products will be evaluated for physical properties to determine the effect of fumigants on the baking performance of flour for household use and on eating quality characteristics of products made from the flours. Flour performance is being measured by oil-binding capacity, amylase activity, mixograph values, and pH. Quality of doughs is being evaluated by Kramer shear values, compressibility, recoil, and pH. Baked bread and baked roll qualities are being assessed by volume, compressibility, Kramer shear, Warner-Bratzler shear, pH, and color. Eating quality characteristics, odor, tenderness, evenness of grain, and flavor of the baked products are being scored by a trained taste panel. Measurements of flour performance and quality will be related to fumigant treatment and residues. The Market Quality Research Division is cooperating in the storage, fumigation, and milling of the wheat, in determining the pesticide residues in milled flour and in making commercial baking and physical dough tests.

## B. Tables of Food Composition

1. B-vitamins and trace elements in foods. Compilation of data is well underway for the following B-vitamins--pantothenic acid, vitamin B<sub>6</sub> and vitamin B<sub>12</sub>, and has been initiated for several trace elements. Data for these nutrients were not in the 1963 revision of Agriculture Handbook No. 8



"Composition of Foods...raw, processed, prepared." Special attention is being given to compiling data on cobalt, copper, manganese, molybdenum, selenium, and zinc.

2. Nutritive values of retail and household units of food. A table is being developed that will give nutritive values for many of the foods in Handbook No. 8 in terms of market units of the items as usually purchased and in terms of household measures of prepared foods. The data needed to prepare this supplement to the 1963 edition of the Handbook are being obtained through consultation with representatives of industry and of Consumer and Marketing Service as well as through observation of products offered for sale.

3. Special services. Research findings compiled from the world's literature on the nutritive value of foods continue to be in constant demand as background material for dealing with a wide variety of problems. Technical assistance was given in the formulation of a statement on nitrogen conversion factors for the Protein Committee of the Food and Agriculture Organization of the United Nations, in the development of policy and guidance for the Food for Peace Program, and in the development of educational materials on the so-called convenience foods for the use of dietitians in teaching diabetics. Information on the composition of specific foods or groups of foods was also given to research teams conducting dietary surveys, to welfare workers and to agencies of the Federal Government such as the Federal Trade Commission, the Food and Drug Administration, and the National Institutes of Health. Within USDA, information was provided for revising several tables in Statistical Bulletin No. 362, Conversion Factors and Weights and Measures for Agricultural Commodities and Their Products, issued in June 1965.

#### C. Nutritional Evaluation of Wheat

The nutritional value of wheat protein and wheat starch is under investigation under research contracts and at Beltsville. At Michigan State University of Agriculture and Applied Science at East Lansing, 12 healthy young men were maintained for 50 days on a controlled diet providing 66.6 gm protein per day of which over 90% was furnished by wheat products and the remainder by fruit and vegetables. The young men remained in good physical condition and were in positive nitrogen balance during the study, an indication that wheat protein is adequate for the needs of young adult men. Most of the constituents of the blood which were included in the analyses remained within normal limits, but the level of urea dropped to about half the normal amount during the dietary period. These findings were presented at the Canadian-U.S. Conference on Nutrition, Toronto, Canada, September 1964. Additional data on amino acids and lipids will be forthcoming.

At the Agricultural and Technical College of North Carolina at Greensboro, a study was initiated to investigate the response of 13 young men to a diet with an appreciably lower protein level than the Michigan study and in which wheat provides 75% of the protein. This diet is being compared to three similar diets in which 20% of the wheat protein is replaced by protein from each of pinto beans, rice, and peanut butter. Data are to be obtained on nitrogen and mineral balance, intake and output of selected vitamins, and on the serum levels of amino acids, total cholesterol, phospholipids, glycerides, and total lipids.

At Beltsville, a study was initiated on the metabolic response of 10 young women to controlled diets in which approximately 85% of the carbohydrate is provided either by wheat starch or by sucrose. Response is being studied in terms of intake and output of nitrogen, fat, and selected minerals, blood serum protein components, blood serum enzymes, blood serum lipid levels, and the fatty acid patterns in the cholesterol, phospholipid, and glyceride fractions of the blood serum.

Research under PL 480 (Hong Kong) was initiated to study the effect on growth and other nutritional indices of children when a significant portion, about 50 percent, of the rice in the diet is replaced with wheat and when their dietary intake of selected nutrients is increased. Approximately 270 children between the ages of 7 and 16 years were studied through one year at an orphanage in Hong Kong. Wheat in the form of bulgur and noodles replaced half the amount of rice in the diets of about 135 children and 135 other children served as controls on the usual rice diet. A mineral-vitamin supplement and a protein supplement were given to a portion of each group. Selected anthropometric and biochemical measurements of nutritional status were made of each child at the beginning of the study and 6 months and 12 months later. Evaluation of the changes in height and weight at the end of one year indicate that there were no significant differences between the groups of children who received all rice as the chief cereal and those who received half of the cereal supply in the form of rice and half as wheat. Results of the biochemical tests and X-rays of the wrists are not yet available.

A project has been initiated to determine the extent of utilization of amide nitrogen and amino acids from wheat for protein biosynthesis and maintenance of tissue protein. Radioisotopes will be employed in these studies as well as the usual criteria for determining nitrogen utilization.

A project has been initiated also to determine whether wheat which has been exposed to one or to multiple fumigations with methyl bromide can cause any physiological disturbance when the wheat is a component of a diet which supplies iodine at marginal levels. The measurements to be made will include those indicative of possible metabolic disturbances that would

reflect the result of a bromide-iodine imbalance on thyroid function. The Market Quality Research Division has arranged for the purchase of the wheat and will treat, store, and analyze the wheat for fumigant residues. Wheat from the same source is being used for studies on the tocopherol and B-vitamin content of the grain and for investigations on baking performance and palatability.

#### D. Food Consumption and Dietary Appraisal

1. 1965 nationwide survey. A nationwide survey designed to provide information on the food consumption and dietary levels of people in the United States is now underway. Data on the kinds and quantities of food used during one week were collected under contract from more than 7,500 representative U.S. households between April 7 and July 3. Similar data will be obtained from 2,500 households each in the summer and fall of 1965 and the winter of 1966. Information on the food eaten both at home and away from home during one day was provided by 13,000 individuals who were members of the families providing information on household food consumption during the spring of 1965. The study was designed to provide data for the four Census regions for farm, rural nonfarm, and urban populations for the year as a whole and for the four seasons.

Detailed tabulation plans have been drawn up that will provide for a series of volumes on the household data obtained in the spring of 1965 similar to those published for the 1955 survey. Other tabulation plans will provide information on (1) the 12-month period April 1965-March 1966 and for the four seasons, (2) the relationship between the money value of household food and its dietary adequacy, and (3) the intake of both food and nutrients of individuals by age and sex.

2. Effects of food distribution programs on diets of needy families. Analyses of data from studies in Detroit, Michigan, and Fayette County, Pennsylvania, conducted to obtain information on the effectiveness of the Food Donation and Food Stamp Programs showed the following. (1) Many of the families participating in the Food Donation Program failed to participate in the Food Stamp Program when it replaced the Donation Program. The families who did participate in the Food Stamp Program were usually those of younger homemakers with more formal education, more young children, and lower incomes for family size. (2) Calcium and ascorbic acid were the nutrients which increased most when the Food Stamp Program replaced the Food Donation Program. They were also the nutrients in which diets were most limited (according to the National Research Council allowances) under both the Food Stamp Program and the Food Donation Program. (3) The overall quality of diets was better under the Food Stamp Program than under the food Donation Program. (4) Under the Food Stamp Program participants received coupons which would purchase food worth considerably more in money than the foods received under the Food Donation Program--approximately 135 percent more in Detroit and 65 percent more in rural Fayette County. They were also able to make their own selections.



A study of families participating in the Food Donation Program in Baltimore showed that little or no dietary improvement occurred when the number of donated foods was increased from five to eight. Families cut back on their food purchases, apparently using the funds thus made available for other needs.

3. Food consumption of the rural population in Spain (PL 480 Research). A 1964 survey of the food consumption of the rural population in Spain, conducted by the Spanish Ministry of Commerce under the cooperative sponsorship of the Economic Research Service and the Agricultural Research Service, using PL 480 funds, showed the percentage of income spent for food was much higher than in the U.S. However, the nutritive content of the diet was considerably lower than that of rural families of the U.S. For example, the average amount of calcium in the diet was about 0.5 g. per person per day compared to 1.2 g. in the diets of rural families in the U.S. in 1955. The percentage of calories from fat was 31 percent compared to 41 percent in the diets of rural families in the U.S. Although a wide variety of foods was used, a large share of the diet was supplied by bread, potatoes, dried beans, milk, olive oil, and wine. Tabulation of data from a second survey conducted in the early months of 1965 is in progress.

4. Nutritive value of national food supply. The revision of estimates of the food energy, protein, fat, carbohydrate, calcium, phosphorus, iron, vitamin A, thiamine, riboflavin, niacin, and ascorbic acid content of the per capita food supply from 1909 to the present has been completed. The revised figures incorporate newest estimates of per capita food consumption developed by the Economic Research Service, revised food composition data from Agriculture Handbook No. 8, and new information on the nutrients added to foods by enrichment and fortification. The revised estimates and tables showing the contribution of major food groups to the total supply of each nutrient for selected years were published in Chapter 5 of Statistical Bulletin No. 364, "U. S. Food Consumption--Sources of Data and Trends," Economic Research Service, June 1965.

The estimates for nutrients together with the per capita food quantities on which they are based are extremely useful in studying dietary trends.

5. Household practices in home freezer management. Field work has been completed for the study of management practices of owners of home freezers. The information was obtained from 200 urban families in Fort Wayne, Indiana, and 200 farm families in the surrounding area. Plans made for tabulating the data will make it possible to describe for groups of families of homogeneous economic characteristics, the types of freezers owned, values attributed to freezer ownership, length of time foods are stored in freezers, and other freezer management practices.

6. Support for food and nutrition programs. The compiling and interpreting of research-based information on nutrition for application to problems of food selection and food use is continuing. The information so developed serves as a basis for assistance to many groups both within and outside the Department. For example, technical advice and guidance were given during the year to the School Lunch Division, Consumer and Marketing Service, in revision of two publications designed to help improve the nutritional quality of school lunches. Also assistance was given to the Office of Economic Opportunity in the preparation of "Nutrition Guidelines" for the Project Head Start Centers Feeding Program.

Publication of Nutrition Program News and participation in the Interagency Committee on Nutrition Education, for which CFE furnishes the secretariat, are continuing as a means of coordinating and strengthening nutrition programs in general. A noteworthy accomplishment was the development by the Committee of four basic nutrition concepts to be used as guidelines for program planning and curricular development in nutrition education.

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### III. MARKETING AND ECONOMIC RESEARCH

#### GRAIN - MARKET QUALITY

Market Quality Research Division, ARS

Problem. Grain and cereal products are subject to damage or deterioration in quality while in the marketing channels through normal metabolic changes, by the action of microorganisms, and by the attack of more than 50 species of stored-product insects. This deterioration affects the grade and price received, the end use, and the wholesomeness of the product. Its effect may be conspicuously blatant, or insidiously hidden; may result in the destruction of nutrient values, or insects may render it esthetically unacceptable. Research is needed not only to protect this multibillion dollar crop from evident deterioration but from the hidden damage as well. Surplus crops and longer storage periods make once accepted standard procedures obsolete. New methods of protection from insects and disease, of identifying and measuring quality changes, and means of maintaining quality over longer periods of time are some of the problems requiring attention.

#### USDA PROGRAM

The Department has a continuing program involving chemists, engineers, and plant pathologists in basic and applied research on the quality evaluation, quality maintenance, and development of objective methods for quality evaluation of cereal grains. The research is conducted at Beltsville, Maryland, and also by cooperative agreement with Purdue University, Lafayette, Indiana, and by research contract with the University of Arkansas, Fayetteville, Arkansas. The program includes the following foreign projects under PL 480: a grant to the Israel Institute of Technology, Haifa, Israel, provides for a study to develop tests for nutritive value of cereal grains and feeds. Its duration is 4 years, 1960-1964, and involves PL 480 funds equivalent to \$103,785 in Israeli pounds.

A grant to the Agricultural Higher School, Poznam, Poland, provides for a study of the effect of microflora of wheat flour on its stability, biochemical, and technological properties. Its duration is 4 years, 1961-1965, and involves \$13,091 equivalent in Polish zlotys.

The Federal scientific effort devoted to research in this program totals 6 professionals divided as follows: quality maintenance and evaluation 5; and contract research 1.0.

There is also a continuing program involving entomologists and chemists engaged in basic and applied research on problems of insect infestation, damage, and contamination of grains and cereal products in the marketing channels.

The activities in Manhattan, Kansas, and Tifton, Georgia, are in cooperation with the respective State Agricultural Experiment Stations. The work at Manhattan, Tifton, Savannah, and Watseka, Illinois, is in cooperation with the Agricultural Stabilization and Conservation Service. During the first half of the fiscal year the equivalent of 0.7 professional man-year of effort distributed between Manhattan, Savannah, and Watseka was supported by CCC funds. At the end of December 1964 the experimental binsite at Watseka was closed down after having been supported for several years by CCC funds. The entomologist was transferred to Manhattan and assigned to research supported by regular funds. The CCC also makes available various commodities and storage facilities for experimental use, thus greatly facilitating the program. There is cooperation with the Field Crops and Animal Products Research Branch of this Division in studies relating to quality maintenance during storage, and on quality evaluation of grains and cereal products; and with engineers of the Transportation and Facilities Research Division on matters relating to storage structures and grain aeration. There is cooperation with growers' cooperatives at Manhattan and with various industry groups at all locations. There is also overall cooperation with several State Experiment Stations through participation in Regional Project WM-52, Maintaining Marketability of Stored Grain and Cereal Products Through Insect Control by Methods Leaving No, Or a Minimum Of, Pesticide Residues.

The program was supplemented by eight PL 480 projects as follows:

A grant to the Direzione Generale dell' Alimentazione, Ministry of Agriculture and Forests, Rome, Italy, is for a study of insect infestation in macaroni, noodles, and spaghetti, and of ways to prevent such infestation. It became effective in 1962, continues through December 1965, and involves P.L. 480 funds with a \$42,621.90 equivalent in Italian liras.

A grant to the Administration of Agricultural Reserves and Surpluses, Montevideo, Uruguay, is for the study of underground and aboveground storage of corn in airtight silos in relation to maintaining quality and preventing insect infestation during long term storage. It became effective in 1962, continues to May 1967, and involves P.L. 480 funds with a \$73,041.93 equivalent in Uruguayan pesos.

A grant to the Indian Agricultural Research Institute in New Delhi is for a study of the varietal resistance of wheat kernels to damage by the rice weevil and lesser grain borer. It became effective in 1965, continues to February 1968, and involves P.L. 480 funds with a \$15,146 equivalent in Indian rupees.

A grant to the Hebrew University at Jerusalem, Israel, was originally for basic research on the influence of environmental conditions on the population dynamics of the khapra beetle. It was extended two years for further biological investigations. The project became effective in 1961, was extended

through October 1966, and involves P.L. 480 funds with a \$97,123 equivalent in Israeli pounds.

A grant to The Maharaja Sayajirao University of Baroda, India, is for basic research on the physiology of fat metabolism in relation to diapause in the khapra beetle. It became effective in 1965, continues through January 1970, and involves P.L. 480 funds with a \$33,907 equivalent in Indian rupees.

A grant to the Instituto Superiore di Sanita in Rome, Italy, was to study the fate of insecticide residues in wheat during storage, milling, and baking or processing into wheat products. The project was terminated prematurely but excellent information was obtained during its short duration.

A grant to the University of Helsinki, Finland, is to study the effects of pesticides on plant commodities. Part of the study will be on wheat. The project became effective in 1964, will continue through December 1969, and involves P.L. 480 funds with a \$96,441 equivalent in new Finnish finnmaks.

A grant to the Hebrew University in Rehovot, Israel, is to study the effect of ethylene dibromide on farm animals. It became effective in 1961, has been extended to August 1966, and involves funds with a \$177,441.99 equivalent in Israeli pounds.

The Federal scientific effort devoted to research on prevention of insect infestation totals 8.2 professional man-years. Much of the cross-commodity research at Savannah, Georgia, reported in Area 13, "Insect Control in Marketing Channels," is also applicable to the insect problems in grain and grain products.

Line Project MQ 0-0-1 (CCC) was discontinued in December 1964 when CCC financial support was withdrawn. Some phases of the project are continuing under regular funds.

#### PROGRAM OF STATE AGRICULTURAL EXPERIMENT STATIONS

State stations are engaged in both basic and applied research on the market quality of grains. A discussion of the program on insect control is included under Area 13.

Considerable effort is devoted to the varietal and quality improvement of grains. For example, when barley is grown for malting purposes, the malting factors are evaluated to determine what effects various varieties, cultural practices and environments have on malting quality. Twelve States are researching the various phases of quality of cereals. The four Federal regional wheat laboratories are located in four of these States. Both State and Federal researchers cooperate closely on this program. Research on the



quality of soft red winter wheat is coordinated through the NCM-28 regional project, with the Federal laboratory at Wooster, Ohio participating in this regional project along with the Ohio, Indiana and Missouri stations. Basic studies involve the improvement of testing methods, the chemistry of bleaching action on starch, lipid and protein interactions in cake baking tests, and the identification of proteins and amino acids associated with good baking quality. Fractionation of flour, particle size, as well as density and air classification, is being studied with wheat flour.

The quality characteristics of experimental strains of wheat are evaluated as well as changes in quality of wheat associated with time and conditions of storage. The various chemical and physical measures are applied in evaluation of quality factors.

There are about 5.7 professional man-years devoted to research on the market quality of grains.

#### REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

##### A. Objective measurement and evaluation of quality

1. Quality Indicators for Stored Wheat. Selkirk, a hard red spring wheat, and Triumph, a hard red winter wheat, harvested in 1963 were placed in storage at 86° and 50° F. and 12, 14, and 16 percent moisture content. The results show that the wheats stored at 86° F. changed at a faster rate than at 50° F. Also, some of the changes were less pronounced for Triumph than for Selkirk. Fat acidity values generally increased faster and losses in sedimentation values were greatest in the spring variety. This suggests that wheats of different classes do not react alike under the same conditions of storage.

The capacitance-resistance determination, a relatively quick, simple test, and the glutamic acid decarboxylase activity determination (methods for measuring deterioration in wheat) proved to be relatively satisfactory measures for following the changes (losses) in stored wheats. Physical dough tests (mixograph and farinograph) indicated that the mixing tolerance properties, an important consideration to the commercial baker, were improved by storage even for the grain stored at 50° F.

According to the baking tests, there is an indication that Triumph is perhaps stronger, changing less (decreasing) in loaf volume than Selkirk under these conditions of storage. Samples of the highest moisture content (16%) evidenced generally the greatest decline during storage in overall quality, with the 14% moisture samples less and the 12% moisture samples the least.

2. Sampling Research. A cooperative agreement was signed with Cargill, Inc. for use of elevator facilities, and the necessary installations made for evaluating mechanical grain sampling devices. These are now in progress.

3. Test Weight-Flour Yield. Methods of measuring bran content, or bran and germ content, and endosperm content as indicators of the flour-yielding capacity of wheat are being investigated. Three methods for measuring bran content are dye absorption, pigment measurement, and lactic acid digestion. In each of the three methods, the germ content of the wheat kernel is also believed to influence the measurement so that it is actually a measure of the bran and germ content. The lactic acid digestion, which leaves a branny residue, appears to be the most sensitive of the three methods in relation to flour yield. Collection and analysis of data on these three methods is not yet complete.

A small mill, the Brabender Quadramat, is being tested as a possible mechanical indicator of the flour yield from the larger Buhler experimental mill. Preliminary results indicate correlations for Quadramat vs. Buhler yields are better for hard wheats than for soft wheats.

Like last year, differences in milling characteristics among the classes of wheat continue to show up in practically all methods of measuring flour yields.

4. Protein Content of Sorghum and Other Feed Grains. A colorimetric-sulfosalicylic acid test used for measuring protein content of wheat was modified by doubling the sample weight as well as reagent. A significant correlation was obtained between meter readings and Kjeldahl protein on 44 samples of milo.

5. Kernel Hardness of Wheat. This project was recently approved and no report has been received from the investigator.

6. Macaroni and Spaghetti Products. The most satisfactory results have been obtained with two color reaction modifications of the Matweef procedure but the tests showing the most promise are time consuming. Optical density measurements of the sitosteryl palmitate fraction also detect in a relatively satisfactory manner, the presence of the different proportions of common wheat used in flour mixtures and finished macaroni products.

7. Mixed Corn of High and Low Moisture Contents. Eight series of moisture determinations on individual kernels, each involving 140 kernels taken from the same sample, have been made. The results indicate the variation in moisture content of individual kernels found in the unmixed samples.

8. Aflatoxin in Corn. Culture extracts produced by 107 fungi isolated from corn grains were assayed by thin layer chromatography for aflatoxin. Certain isolates of Aspergillus awamori, A. flavus, A. parasiticus, A. ruber, A. wentii, Penicillium citrinum, and P. variable produced aflatoxin. Penicillium cyclopium, P. frequentans, and P. puberulum elaborated this toxin only in trace amounts. Bioassays of extracts from 4 of these fungi showed that only the extract from A. parasiticus was highly toxic.

9. Baking Quality of Wheat. A satisfactory baking quality test is a continuing need for the quality evaluation of wheat. A simple flour-disc test is promising for determining bread baking quality in wheat. The flour-discs were made by pressing a mixture of ground whole wheat and lactic acid in a Carver press. Agtron reflectance readings of the flour-discs gave highly significant correlations with protein, sedimentation, and loaf volume values. Moisture meter correction factors were established for measuring moisture content in various types of wheat treated with diatomaceous earth for hard red spring, hard red winter, soft red winter, and white wheat.

#### B. Quality maintenance in storage

1. Changes in Wheat Due to Insecticides. After storage for one year, wheats treated with Cab-o-sil, Silica Aerogel 68, Perma Guard, and Kenite, showed no further decrease in test weight per bushel and no change in the yield of flour, as compared with the untreated wheat. The malathion-treated wheat was not changed in test weight or yield of flour after storage. The different dust or liquid insecticide materials appear to have no effect on the fat content of the treated and stored samples as compared with the control wheat. Normal cleaning of the grain in preparation for milling removed a large percentage of the dust insecticide materials judged by visual inspection. There were no important differences in chemical quality tests, in physical dough tests, or bread baking characteristics of the wheats between initial treatment and after 1 year of storage compared to the control sample.

2. Changes in Wheat Due to Radiation. Samples of hard red winter wheat treated with 0, 10, 25, 50, 125, and 175 thousand rads of gamma radiation have been stored one year at 75° F. Initial germination values were unchanged by 10 and 25 thousand rad treatments, materially decreased by 50 thousand rads and destroyed by the 125 and 175 rads treatments. The losses in germination were less for the 10 thousand rad treated wheat than the other treated samples after 1 year of storage. There were significant initial losses in sedimentation values with increasing levels of radiation, but no additional losses in sedimentation after 1 year of storage. Irradiation treatment had no effect on initial fat acidity but storage for 1 year increased the values somewhat. Enzyme activities, as indicated by the glutamic acid decarboxylase values were unchanged by increasing initial dosages of irradiation but showed marked decreases with time of storage. The capacitance resistance determination



a rapid test for deterioration in corn, showed no changes in the wheats when initially treated but increased gradually during the 1 year storage period. The treatments tended to slow down desirable physical changes in dough properties, but had no effect on loaf volume. Dosages of 125 and 175 thousand rads produced distinct off odors (burned, charred, or decayed) in the hot breads but these were not evident in the cold loaves. A trained judging panel found no significant difference in the taste of the breads from the irradiated and non-irradiated wheat.

3. Microflora of Wheat. Conditioned wheat (cleaned, washed, and heat treated) produced a higher yield of flour than either the cleaned or washed grain. The flour from the conditioned wheat spoiled more rapidly on prolonged storage than the flour from untreated washed grain. Fat acidity values of the flour were highest in the untreated and washed grain and lowest in the conditioned wheat. Flour stored at 20° C. and 90% relative humidity decreased in fat acidity after 8 months' storage, suggesting complete spoilage of the flour and destruction of the fats and free fatty acids due apparently to molds. The bread-baking characteristics in terms of loaf volume, have generally improved, for the flours stored under various conditions except for storage at the highest temperature (20° C) and relative humidity (90%). Organoleptic evaluations, surprisingly, showed development of off odors and flavors in the bread from flour stored 10 to 12 months at the low temperature of 3° C.

4. Long-Term Storage of Wheat. Nine samples of Marquis, a spring wheat grown under irrigation from 1921 to 1929, and four samples of Kanred, a winter wheat grown without irrigation during a like period of years, were stored 35 to 43 years at Fort Collins, Colorado, in bags in a dry unheated room. The germination of the wheats decreased progressively with age but Kanred lost viability somewhat faster than Marquis harvested in the same years. There was no change in protein content during storage but the ash content of the flour averaged consistently higher than when the original tests were made (1938). Thiamine content was generally comparable to that of freshly harvested wheat of the same varieties. There was a definite and fairly uniform increase in fat acidity during storage. Sedimentation values determined for the first time in 1964 were low compared to recently harvested wheat of like variety. Dough mixing properties (by the farinograph) for the 35 to 43 year old wheat were remarkably strong with Marquis somewhat superior to Kanred. The quality of the bread decreased somewhat with greatest losses in bread grain and texture.

Tests of these stored wheats have been made periodically. This recent study is being prepared for publication and will constitute the fifth paper in the series on this subject.

## C. Prevention of insect infestation

1. Nonpesticidal Control Methods. Small-bin studies with varied dosages of inert dusts showed that 2 diatomaceous earths were highly superior to 2 silica aerogels over a period of 15 months. The diatomaceous earths were also superior to the standard dosage of malathion.

In a field test on corn in Illinois, malathion, 2 diatomaceous earths, and 2 silica aerogels all continue to give a high degree of protection against insect infestation 21 months after treatment. At the end of 21 months of a field test with corn in Georgia, a diatomaceous earth and a silica aerogel continue to be comparable with malathion in protecting against insect infestation. In laboratory bioassay tests of corn from the field test, the inert dusts gradually decreased in toxicity and repellency as the length of storage increased. In these field tests the flow rate of grain through grain handling equipment was greatly reduced in the dust treatments and test weight was lowered several pounds per bushel. It was also exceedingly difficult to insert probes into the dust-treated grain for inspection and sampling. None of these difficulties resulted from the malathion treatment.

Seventy-one test crosses of corn from the Crops Research Division were tested for repellency to the rice weevil, using a common hybrid, Coker 71, as the reference check. Thirty-three of the test crosses showed repellency, six rating above 55 percent.

Some rice weevil, granary weevil, and lesser grain borer adults and eggs were able to survive exposure in laboratory tests to extremes of vacuum, pressure, or temperature exceeding those used in pasta manufacturing processes. Other investigations still indicate that insect infestation of pasta occurs after manufacture. Examination of almost 18,000 weevil galleries in "camolata" pasta from all over Italy showed that the eggs had been deposited in the pasta not only after it had been ejected from the dies but also after final processing.

Corn was sampled from aboveground hermetic silos after storage for one year in tests in Uruguay. The corn was in good condition and no insects were present. Air samples from within the bins contained 27 to 46 percent of carbon dioxide and only 0.18 to 0.5 percent of oxygen.

2. Biology, Ecology, and Physiology. A seasonal cyclic variation in composition of air in 3,250-bushel metal bins of wheat was found in Kansas. There is a minor build-up of  $\text{CO}_2$ , reaching a peak of 0.16 percent in October. As the grain cools, the concentration decreases slowly, becoming lowest in April. As the grain warms the  $\text{CO}_2$  again increases to a peak in October.

Preconditioning adult flour beetles with CO<sub>2</sub> or N<sub>2</sub> to increase susceptibility to fumigation was more effective at 30° C. than at 20° or 40° C. Confused flour beetles secreted large amounts of paraquinones during recovery from CO<sub>2</sub> or N<sub>2</sub> preconditioning, and these compounds accumulated in toxic amounts if not removed by active ventilation. Insects were most susceptible to low concentrations of methyl bromide fumigant while in a condition of depressed rather than high respiration rate. It is assumed the detoxification of methyl bromide by the insect was slower at low metabolic levels. Preconditioning did not affect the insect's susceptibility to chloroform as a fumigant when exposure was at either a high or low rate of respiration.

Rice weevils appear to prefer clean wheat to that with high concentrations of dockage. But when immature stages were left in the wheat to continue their development more progeny were found in wheat with greater amounts of dockage.

When 1 to 4 pairs of adult khapra beetles were placed in 3.5 grams of wheat, the greatest number of larvae was produced by the 4 pairs but the greatest number of pupae resulted from the single pair. The higher the population density the greater was larval mortality and the longer was the larval developmental period for females. Wheat kernels from a khapra beetle culture appear to contain substances that cause mortality and delayed development of larvae reared on that food. Rinsing partly eaten grain in water or ether appeared to remove the adverse factor, but when insects were reared on the ether-rinsed grain an unexpectedly high proportion, 77 percent, of the resulting beetles were females. It appears that a petrol-ether solvent, but not water, removes from khapra beetle excreta a factor that inhibits pupation and induces diapause in larvae. Khapra beetle larvae were strongly attracted to an extract of larvae of the same species in olfactometer tests, and there are indications the attractant is rather volatile. Khapra beetle larvae were not attracted by extracts of four other species of larvae.

3. Improved Pesticidal Control. Applications of 5 p.p.m. of diazinon or fenthion to grain, then aged for one year, were equal or superior to malathion at 10 p.p.m. against test insects in the laboratory. Diazinon or fenthion at 10 and 20 p.p.m. and aged one year were completely effective against all species of test insects. Carbaryl was found to have definite specificity for control of the lesser grain borer.

In tests of fumigants for use behind the linings of empty rail cars, a mixture containing 70 percent of ethylene dibromide and 30 percent of methyl bromide was better than one containing 80 percent of carbon tetrachloride and 20 percent of carbon disulfide. Laboratory tests with phosphine show a higher concentration-x-time factor is required against flat grain beetles than other common stored-grain insects.



In studies of physical behavior and distribution of liquid grain fumigants in corn, cold weather adversely affected the mixture containing 75 percent of ethylene dichloride (EDC) and 25 percent of carbon tetrachloride ( $\text{CCl}_4$ ). Volatilization of the EDC was delayed and distribution was limited to the upper half of the bin. Distribution of a mixture containing 80 percent of  $\text{CCl}_4$  and 20 percent of carbon disulfide was not adversely affected by cool grain. Recovery ratios of the 80:20 mixture throughout warm or cool grain were similar to applied ratios. There was little similarity to applied ratios in the case of the 75:25 mixture.

4. Fate and Effects of Pesticide Residues. In PL 480 studies in Italy, malathion residues decreased slowest during storage in American Hard Red Winter wheat, intermediate on Italian durum, and most rapidly on Italian soft wheat. Residues were not detectable in the latter after 12 months' storage. When treated wheat was milled, very little malathion came through into the white flour. Most of it was in the bran and shorts. White flour contained only about 0.5 p.p.m. when milled from American Hard Red Winter wheat after being treated with 10 p.p.m. of malathion and stored for 1 year. The residue was negligible in flour from Italian durum and undetectable in that from Italian soft wheat. Bread contained only 8 to 16 percent of the malathion present in flour from which it was baked. Uncooked pasta contained only about 10 percent of the malathion present in flour from which it was manufactured, and no residue was detectable in cooked pasta. These results are of particular importance to the U. S. grain trade because European Common Market countries have been critical of malathion residues in our wheat. This report from one of their own countries says "... the use of malathion in the storage of wheat can be considered safe for the consumer."

In similar studies with methoxychlor, wheat treated with 27 p.p.m. lost only 0.2 to 0.4 p.p.m. during 6 months of storage. White flour contained 9 p.p.m., shorts 70, and bran 100.

5. Toxicology. By use of a newly-developed gas-liquid-chromatographic analytical method, EDB was found to be the only volatile compound remaining in materials treated with this fumigant. This indicates the absence of volatile reaction or degradation products. The largest amount of residual bromine was in lentils, which contained the largest amount of protein of the feeds tested. Addition of ascorbic acid or methionine to the diet of laying hens did not counteract the effects of EDB in reducing egg size and number. The cause of EDB effect on egg size does not appear to be directly hormonal. Prolonged feeding of EDB to mature cows did not affect conception and gestation, but in young heifers given EDB from birth, conception was delayed. Young cattle were affected by all doses higher than 2 mg./kg. The principal effect of EDB on ruminants appears to be on bull spermatozoa. The chief detoxication product of EDB in rat urine was identified as S(Beta-hydroxyethyl)-N-acetyl-cysteine. Feeding of EDB-treated mash to mature cocks did not affect volume of semen, sperm motility, or sperm concentration

after 2, 4, or 6 weeks of treatment. The histological effects of feeding EDB-treated food from calthood to full maturity were shown to be: (1) spermatogenesis was markedly reduced and, in many cases, completely ceased, (2) Sertoli cells seemed normal, while spermatogonia were in the resting stage, (3) most of the lumina of the tubules were empty or else filled with cell debris, (4) Leydig cells were normal, (5) in the caput and corpus epididymis, elongation of the columnar cells was found, which caused the epithelial lining to protrude into the lumen of the ductus, and (6) thickened connective tissue was characteristic all along the epididymis.

#### PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

##### Objective Measurement and Evaluation of Quality

Hart, J. R., and C. Golumbic. 1965. The use of electronic moisture meters for determining the moisture content of seeds. Preprint, 14th International Seed Testing Congress, Munich, May.

Baker, Doris, C. C. Fifield, and T. F. Hartsing. 1965. Factors related to the flour-yielding capacity of wheat. The Northwestern Miller, February.

##### Quality Maintenance in Storage

Hart, J. R. 1964. Hysteresis effects in mixtures of wheats taken from the same sample but having different moisture contents. Cereal Chemistry, Vol. 41, No. 5, September.

##### Prevention of Insect Infestation

Amir, D., and R. Volcani. 1965. Effect of dietary ethylene dibromide on bull semen. Nature 206(4979): 99-100.

Dal Monte, Gino. 1965. Studi sull' origine delle infestazioni di puncteruoli nelle paste alimentari. (Studies on the origin of weevil infestations in pasta.) Agricoltura, Vol. 4, 9 pp.

**RICE - MARKET QUALITY**  
**Market Quality Research Division, ARS**

Problem. Harvested rice is subject to damage or deterioration in quality while in marketing channels through normal metabolic changes, by the action of disease organisms, and by insect infestation. There is need for developing more effective ways of preventing insect infestation during storage, handling, processing, packaging, and transportation of rice. Attention must be given to finding control methods that will minimize or eliminate pesticide residue hazards. To maintain the quality of rice, more precise information is needed on the changes that occur in handling, storage, and transportation. To insure uniform and standardized products and more equitable prices to all concerned, new and improved procedures for measuring quality factors must be developed for use in inspection, grading, and standardization operations.

**USDA PROGRAM**

The Department has a continuing program involving engineers, chemists, and plant pathologists in basic and applied research on the quality evaluation and quality maintenance of rice. This work is located at College Station, Texas, in cooperation with the Texas Agricultural Experiment Station.

The Federal scientific effort in this area totals 2.5 professional man-years: quality evaluation 0.5 and quality maintenance 2.0.

A grant to the Department of Plant Chemistry, Valencia, Spain, provides for a study on storage changes in milled rice and their relation to market quality. Its duration is for 4 years, 1964-1968, and involves P.L. 480 funds with a \$62,479 equivalent in Spanish pesetas.

There is also a continuing program involving entomologists and chemists engaged in basic and applied research on the prevention of insect infestation in rice in the marketing channels, headquartered at Fresno, California. The Federal effort of about 2 professional man-years was temporarily diverted during this reporting period to emergency research on the effects of gamma irradiation on stored-product insects, which is also pertinent to the problems of insect infestation in stored rice. Much of the cross-commodity research at Savannah, Georgia, reported in Area 13, "Insect Control in Marketing Channels," is also applicable to the problems in stored rice.



## PROGRAM OF STATE AGRICULTURAL EXPERIMENT STATIONS

State stations maintain a continuing but very modest program of research related to the market quality of rice. It involves evaluation of new rice varieties and lines for specific qualities through cooperation with the Regional Rice Quality Laboratory. Basic chemical and physical data relative to the quality of the rice and its protein, lipid, and starch components is sought. The variation in these constituents among several varieties and in relation to maturity at harvest is also investigated. Another study is concerned with the effects of infrared drying of rough rice on the quality of the milled rice.

The total State scientific effort devoted to market quality research on rice is 1.6 professional man years.

### REPORT OF PROGRESS OF USDA AND COOPERATIVE PROGRAMS

#### A. Objective measurement and evaluation of quality

1. Degree of Milling of Rice. Light absorption and reflection properties of white and parboiled milled rice showed: (1) that the most effective method of measuring degree of milling is a measurement of the ratio of light transmittance at 850  $m\mu$  to that at 660  $m\mu$  and (2) color and degree of parboiling can best be measured by reflected light--the tristimulus value called "lightness." Measurements of degree of parboiling have shown that when samples are carefully graded by a number of inspectors and assigned numerical values representing true differences, the correlation between meter reading and visual grade is 0.935. These results substantiate the accuracy of the experimental lightness meter in predicting both color of white milled rice and degree of parboiling of parboiled milled rice. A commercial instrument manufacturer is now constructing the combination rice meter.

#### B. Quality maintenance in handling and packaging

1. Microbiological, Chemical, and Physical Deterioration of Rough Rice. Infrared drying of Century Patna rough rice lowered the equilibrium moisture

content during storage at 75% and 85% relative humidity an average of 0.3 - 0.4% over a period of 6 months. The rate of infection by storage fungi was significantly reduced by treatment with sodium propionate for 6 months in samples stored at 75% relative humidity and 4 months in samples stored at 85% relative humidity. Sodium propionate suppressed infection by Aspergillus candidus but did not control A. glaucus group spp.

Preharvest infection of Belle Patna rough rice by Helminthosporium oryzae did not materially affect the subsequent mycofloral succession during storage over a period of 385 days under marginal storage conditions (75% relative humidity at 30°C). Species of the A. restrictus series became the pre-dominant fungi, invading both H. oryzae infected kernels and fungus-free kernels. After 385 days, A. restrictus spp. were isolated from 90%+ of the kernels. Although total yields of brown and milled rice did not change significantly during the storage period, head rice yields decreased 2.7% in the lot with a 2% initial infection by H. oryzae in comparison to 4.4% in the lot with a 64% initial H. oryzae infection. There was also a decrease in the number of visually detectable damaged (discolored) kernels in brown and head rice in both lots.

The incidence of storage fungi in 207 lots of commercial rough rice of the 1964 crop from the 4-state southern rice area was determined and is given as the percent of lots from which the various major groups of storage fungi were isolated from a 100 kernel sample per lot: (1) A. flavus-oryzae group, 98%; (2) A. glaucus group, 95%; (3) A. niger group, 45%; (4) A. candidus group, 37%; (5) other Aspergillus spp., 12%; and Penicillium spp., 67%.

2. Mycotoxins in Rice. In a total of 729 samples of rough, brown, and milled rice assayed, 0.27% (2 samples) contained aflatoxins in concentrations greater than 50 ppb. Aflatoxins were not detected in 88.75% of the samples. In 77 randomly selected isolates of Aspergillus flavus-oryzae, twenty-two produced less than 5 ppb aflatoxin on sterile rough rice incubated 10 days at 30°C. The production by the remaining 45 isolates ranged from 5 ppb to a maximum of more than 500,000 ppb as determined by thin-layer chromatography on silica gel plates. Similarly, measurable amounts (max. 2957 ppb) were produced by 6 of 33 isolates of Penicillium spp. In a test with one isolate of A. flavus, more aflatoxins were produced on a substrate weight basis on ground brown rice than on ground rough rice, ground milled rice, rice polish, and rice bran. Other studies have indicated that rice is equal to peanuts as a substrate for the production of aflatoxins. Significant interactions, expressed in yields of aflatoxins have been observed between strains or isolates of fungi and peanut and rice substrates.

Non-sterile rough rice was inoculated with dry spores of a known high-aflatoxin-producing strain of A. flavus and stored at 30°C in relative humidities ranging from 70 to 100% for periods up to 4 weeks. The toxin concentration in the rice exceeded 50 ppb after one week in storage in relative humidities of 85% and above. At 80% and below, only traces of aflatoxins were detected during the entire storage period. The maximum concentration detected was in excess of 40,000 ppb after 2 weeks in 100% relative humidity.

C. Quality maintenance in storage

1. Storage Changes in Milled Rice. Initial experiments have indicated that changes in the molecular characteristics of the amylopectins and in starch content may be associated with aging and related changes in quality.

D. Prevention of insect infestation

1. Nonpesticidal Control Methods. During studies on the effect of infrared drying on insects in rice, it was found that the variety of rice may influence the results. Final temperatures attained by Pearl, Calrose, and Belle Patna varieties varied after treatment with similar dosages of infrared radiation. This point is being investigated further.

A brief survey of the rice storage and milling industry operations in the Sacramento Valley of California revealed a general awareness of the importance of sanitation as a factor in preventing insect infestation. Carrying out good sanitation programs is apparently helping to keep insect infestations at a low level. This reduces the need for chemical control, which in turn helps to avoid pesticide residues.

2. Improved Pesticidal Control. American Cyanamid No. 47,300 was the only one of 5 candidate compounds found promising in preliminary laboratory evaluations of potential protectants for stored rice. It is receiving further testing.



The previously mentioned survey of the rice industry in California also revealed that pesticides were being used on a preventive maintenance basis to very good advantage. Such preventive use helps minimize more extensive pesticide applications that would otherwise be required as control measures.

#### PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

##### Objective Measurement and Evaluation of Quality

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Johnson, R. M. 1965. Light reflectance meter measures degree of milling and parboiling of parboiled rice. Cereal Chemistry 42:167-174.

##### Quality Maintenance in Storage

Schroeder, Harry W. 1964. Sodium propionate and infrared drying for control of fungi infecting rough rice (Oryzae sativa) Phytopathology 54:858-862.

##### Prevention of Insect Infestation

Tilton, Elvin W., and Robert R. Cogburn. 1964. Laboratory evaluation of Bayer 29493 used for the protection of rough rice against insect attack Proceedings Rice Technical Working Group, Davis, California.

**FEED AND SEED - MARKET QUALITY**  
**Market Quality Research Division, ARS**

Problem. Many methods of determining seed quality currently in use require too many man-hours, impose tedious work on the analyst, are incapable of high degrees of standardization, and do not provide accurate indexes of quality. Practical methods are needed for determination of such quality factors as mechanical purity, genetic purity, germination, vigor, weed seed content, and infection with disease organisms. The deleterious effects of high temperatures and relative humidities on stored seed are well known but little is known about the part played by storage molds, especially the minimum temperature - relative humidity combinations under which the storage molds survive in stored seed. There is urgent need to increase basic research which would serve as a basis for developing more practical methods of determining seed quality and for recommending improved practices of storing seed.

**USDA PROGRAM**

The Department has a continuing long-time program on seed research involving botanists, plant physiologists, plant pathologists, engineers, and chemists engaged in both basic and applied research on quality evaluation and quality maintenance of seed. This research is conducted at Beltsville, Maryland, and College Station, Texas, and by research contract with the Oregon Agricultural Experiment Station.

A P.L. 480 grant with the Instituto Biologico, Sao Paulo, Brazil, provides for a study of substrate moisture levels for germination testing of agricultural seeds. The project runs from 1962 to 1967 and involves \$31,016 equivalent in Brazilian cruzieros.

A P.L. 480 grant with Rijksproef-station, Wageningen, Netherlands, provides for a study of the health condition of seeds in commercial channels and development of methods suitable for routine testing for seedborne organisms. The duration of the project is 5 years, beginning 1963, and the total grant in Dutch guilders is the equivalent of \$55,777.

A P.L. 480 grant with Samenprufstelle, Munster, Germany, provides for a study of the biological and environmental factors affecting the physiological maturity of grass seeds. The duration of the project is 3 years, beginning April 1965, and the total grant in German marks is the equivalent of \$31,775.

A. P. L. 480 grant with Forschungsgemeinschaft fur Saatgutforschung, Reutligen, Germany, provides for a study of methods for maintaining the germination of seeds in storage and in trade channels. The project has a duration of 5 years, beginning April 1965, and the grant in German marks is the equivalent of \$52,338.

A P. L. 480 grant with Agricultural Research Station, Beit Dagen, Israel, provides for research to find a satisfactory invisible marker of seeds in commercial channels and for research purposes. The project runs for 3 years, beginning February 1965, and the total grant in Israeli pounds is the equivalent of \$45,640.

A P. L. 480 grant with Indian Agricultural Research Institute, New Delhi, India, provides for a study to evaluate the X-ray technique for detecting empty seeds in purity testing and for determining seed viability. Its duration is 5 years beginning November 1964 and the total grant in Indian rupees is the equivalent of \$37,464.

A P. L. 480 grant with the Israel Institute of Technology, Haifa, Israel, provides for a study to develop tests for nutritive value of cereal grains and feeds. Its duration is 4 years, 1960-1964, but an extension of two additional years has been approved. The project involves an expenditure of \$31,016 equivalent in Israeli pounds for the original 4-year period.

The Federal scientific effort devoted to research in this area totals 8.0 man-years of which 2 man-years are by research contract. A project on seed moisture determination was terminated

#### PROGRAM OF STATE AGRICULTURAL EXPERIMENT STATION

Scientists at the State stations have engaged in the study of the market quality of feed and seeds for many years. The program is continuing and involves both basic and applied research.

Much of the research on feed quality is conducted in conjunction with the extensive program in animal nutrition. Quality of feed studies relate to determining keeping quality of feeds as influenced by methods of harvesting, storing, and handling of livestock feeds. Mechanical developments facilitating the mechanization of harvesting and handling feeds have led to consideration of alternate feed handling and storage methods and evaluation of their effects on feed quality. Attempts to store feedstuffs with a minimum loss of quality and nutritive value have led to study and development of routine methods for evaluation of vitamins A and D in feeds. Biological changes occur in feeds during storage and the relationship of these changes is related to deterioration of computer formulated, least cost feeds with quality, and nutritive value considered as important factors.



Seed quality research involves physiological studies on factors associated with seed deterioration, the sequence of biological changes in storage, storage factors affecting these changes, seedborne microflora, and the use of seed coatings to preserve quality. Testing procedures for germination, vigor, and stand-producing potential are receiving much research effort. Other work is being done on blending seeds for uniform quality.

Two regional projects have been organized by the States to coordinate research on seed problems, including quality considerations. In the Northeastern region under NCM-22, quick methods of determining varietal purity of alfalfa and red clover are sought through both field and laboratory experimentation. In the Western region under WM-35, much attention is given to developing and improving techniques for rapid estimation of such quality factors as viability of seed, germination, and purity.

The total research effort in this area is 20.6 professional man years; of which 1.7 is devoted to feed quality research, and 18.9 professional man years is devoted to study of post harvest physiology of seeds and objective measures of quality in seeds.

#### REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

##### A. Objective measurement and evaluation of quality

1. Mechanization of Purity Analysis. A treated seed inspection station and a vibrator separator were completed and construction specifications and blueprints prepared. An improved subsampler was developed. It appears to be as accurate as the Boerner divider but is self cleaning, thus avoiding accidental contaminations with seeds lodged in hidden areas of the divider. This subsampler has been "field" tested and will be used in future research.

2. Seed Metabolism. Further examination of the ribosome system in the germinating seed has allowed the identification of the polysome (an aggregate of the ribosome) as the component responsible for protein synthesis. The shift from monosome (single ribosome) to polysome

accompanying imbibition, indicates that in this transformation resides the control of protein synthesis by the seed. Analyses by in vivo labeling have further confirmed that all steps prior to arriving at the ribosome as well as those involved in release from the ribosome are not rate-limiting. However, kinetic data from this process do not correlate with the kinetics of the monosome to polysome conversion suggesting a more complex process.

3. Determining the Purity of Certain Grass Seeds. A series of referee tests showed that with Bahiagrass seed: (1) there was slightly less variation in results obtained by the flotation method than by the official method, (2) for any one determination, the standard error was 1.03% for both hand and flotation methods, and (3) the accuracy of the flotation method was slightly influenced by sample purity level.

4. Development of Seed Germinator. Tests with a prototype model of the basic germinator under development have resulted in improvements and are being incorporated into a model that is now under construction and comprise: (a) more reliable temperature controller, (b) higher intensity, more uniform lighting of germinator, (c) reduction in the time required to alternate temperatures and extension of the temperature range, and (d) minimizing overshoot in cooling.

5. Processing Grass Seed for Laboratory Testing. A modified McGill rice sheller has been adapted for shelling grass seeds by replacing the steel fluted roll with one of hard rubber. This machine has shelled orchard-grass, sideoats grama, and little bluestem thus far, with little or no damage to the caryopses. Proper settings of the roll spacing has been determined for these grasses.

6. Seed Vigor. Respiration measurements detected injury caused by gamma-irradiation, heat, or freezing of moist seed in corn, and by chilling injury during imbibition in lima beans. Measurements made two hours after the start of imbibition detected the presence of injury, and in the case of heat-treated corn seeds, the test was a more sensitive index of seed quality than standard germination tests, as judged by field emergence. A general relationship appears to exist between initial respiration rates and subsequent seedling growth of corn.

7. Development of Methods Suitable for Routine Testing of Seed for Seedborne Organisms. The principal finding to date is that certain species of fungi grow faster and produce more easily identifiable colonies in germination substrata kept slightly dry than when left moderately moist or wet. While this did not hold true for all species of fungi, it should be of value to those seed testing stations which test flax seed for pathogenic fungi. It appears likely that the procedure can be used.

8. Verification of Varietal Designations of Crop Seeds. Significant differences in protein banding on the acrylamide gel have been found between groups of varieties; however, these groups with different banding patterns can also be distinguished by morphological characteristics of the seeds.

9. Moisture Levels for Seed Germination. Based on dry weight of the seed, the initial water uptake, expressed as percent of the dry weight of seeds, was found to be essentially the same for rice and corn. Crotalaria and canavalia (swine bean) differed considerably from beans, corn, and rice. Earlier, it was stated that a correlation was found between the weight of seed of different species and water absorption. Additional research failed to confirm the earlier reports.

In general, it was found that abnormal seedlings caused by insufficient moisture recovered and produced normal plants but that most of the other abnormal seedlings failed to do so.

Some workers claim that the principal area through which water passes are the hilum and micropyle. It was found that the micropyle does permit some water to enter the seed but that the hilum is essentially impervious to the passage of water. There is a small band just outside the hilum through which water appears to pass relatively freely.

## B. Quality maintenance in storage.

1. Microbiological Deterioration of Grass Seeds During Marketing. Relative humidity had no deleterious effect on seed germination when the storage temperature did not exceed 10°C or at 20°C and 35% or 55% relative humidity. Storage at 30°C was not harmful at 35% relative humidity but at 55% relative humidity a decrease in germination occurred between 8 and 12 months. Storage at 35°C and 55% relative humidity was deleterious when



the storage period exceeded 4 months, but when stored at 35°C and 75% or 95% relative humidity severe damage occurred after 2 months.

There appeared to be no close correlation between the decline in seed germination and the number of species of storage fungi belonging to the Aspergillus glaucus group that were recovered from the seeds. The fungi in this group tended to decrease in number with time. At relative humidities of 35, 55, and 75 percent, temperature had practically no effect on the relative humidity and seed-moisture-content relationship; however, this was not true at 95 percent relative humidity. Research aimed at production of fungus-free seeds has been initiated. Fungus-free seeds are almost essential as controls in the study of fungus effects on seed deterioration.

2. Gamma Radiation of Seeds. There was great variation in sensitivity to radiation injury among the crop species used. Germination of onion seed was reduced, or the emerging radicles injured at 5 to 10 Krads; whereas, the germination of crimson clover and radish seed was not reduced significantly at a dose of 80 Krads. In general, a low percentage of seeds of each seed lot was injured at the dose found to be the minimum for the species concerned; then injury increased as the dose was increased. There was no evidence that high moisture content of the seed (13.5 percent) protected it against radiation damage but there was an indication that low moisture content (5.0 percent) renders the seed more vulnerable to radiation damage.

Seeds that had been stored at 80 percent relative humidity and 21°C (95 percent relative humidity for peanuts) for a total of 16 months were assayed for the internally-borne storage fungus, Aspergillus amstelodami except that peanuts were assayed for A. flavus. Aspergillus amstelodami apparently died out in both the unirradiated and irradiated samples of radish (1963 crop year) and sorghum (1962 and 1963 crop years). The amount of A. amstelodami on seeds of peanuts, Kentucky bluegrass and onion decreased. In corn, no appreciable decrease was evident but there was a large increase in wheat.

#### PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

##### Objective Measurement and Evaluation of Quality

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MARKETING FACILITIES, EQUIPMENT AND METHODS  
Transportation and Facilities Research Division, ARS

Problem. Differences in varieties of individual field crops and in the environments of producing areas where they are conditioned and stored, together with advancing techniques in cultural and harvesting practices, require new or modified marketing facilities, equipment, and methods. Such changes are essential to the efficient and economical handling, conditioning, and storing of these crops and to maintaining their quality. There is a need for improved designs for facilities based on functional and structural requirements, which will expedite the movement of commodities into, within, and out of the facility. There is also a need for handling and conditioning equipment which will minimize labor and other costs. More knowledge is needed of the relative efficiency of various handling and conditioning methods so that improved or revised methods and equipment can be developed to perform necessary operations.

USDA AND COOPERATIVE PROGRAM

The Department has a long-term program involving engineers engaged in both applied and basic research on, as well as application of known principles to, the solution of problems of handling, storing, and conditioning field crops in marketing channels. Grain aeration and drying research is carried out at Manhattan, Kans., on wheat and grain sorghum and at Lafayette, Ind., on corn, in both laboratory and pilot-scale facilities and in commercial storages; in cooperation with the Agricultural Experiment Stations of respectively Kansas and Purdue University (Indiana); and with grain storage firms. This research is supplemented by contracts with the Shuman Chemical Laboratory, Inc., Battle Ground, Ind., and the Aerophysics Company, Washington, D. C. Research on the design of grain storage structures is conducted at Hyattsville, Md., with field studies providing the basis for selecting capacity and type of operation. Research on the handling of grain in country elevators and terminal storages is conducted at Manhattan, Kans., in cooperation with the Kansas Station, and with various commercial firms; and is supplemented by a research contract with Cargill, Inc., Minneapolis, Minn. Studies of grain storage in CCC bins are conducted at a research bin site at Watseka, Ill. Studies on the handling, drying, aerating, and storing of rice are conducted at Beaumont, Tex., in cooperation with the Texas Agricultural Experiment Station and at commercial facilities in Texas, Arkansas, Louisiana, and Mississippi.

The Federal effort devoted to research in this area during the fiscal year 1965 totaled 7.6 professional man-years, including 3.2 to grain aeration and drying; 0.8 to the design of grain storage structures; 0.8 to the handling of grain in terminal storages; 0.8 to studies of grain storage in CCC bins and 2.0 to the handling, drying, aerating, and storing of rice.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Handling Grain in Commercial Storages

At Manhattan, Kans., a report on unloading boxcars of grain at terminal elevators was completed and submitted for review and clearance for publication. The research report evaluates data on economical methods and equipment for unloading boxcars and shows labor requirement tables and equipment ownership costs. Two types of equipment, the trackmobile and the car puller, were studied for moving boxcars at terminals during the unloading operation. Five types of equipment; mechanical and hydraulic car dumpers, car shakers, self-propelled augers, and power shovels, were studied for unloading boxcars. Annual operating schedules were developed for three capacities of terminals as a basis for establishing efficient and practical combinations of unloading equipment for terminals of different sizes.

In a continuing study, grain temperature data were obtained in a concrete upright storage, 18 feet in diameter and 110 feet high, to determine the effect of ambient air temperatures on the temperature of wheat in static (unturned) storage. Temperatures of the stored wheat were most uniform during the latter part of October and early November; the greatest variations occurred during July, August, and December.

At Hyattsville, Md., a research contract has been negotiated with Cargill, Inc., Minneapolis, Minn., for conducting investigations designed to determine the extent and causes of physical damage (breakage) to grain by equipment used in handling grain into, within, and out of marketing facilities. This work was initiated, in part, because of increasing criticism and concern by foreign buyers regarding the quality of U.S. grain--much of which is attributed to the substantial amount of broken kernels present in the grain. Initially, pilot-scale studies will be conducted using full-scale equipment under simulated conditions to study the effects of grain slingers (trimmers, throwers) used in loading ships, boxcars, bins, and trucks; of grain dropping both by free fall onto concrete, steel, grain, etc., and by fall through spouting; and of elevating grain by belt and bucket elevators at various bucket speeds and loading.

### B. Design of Grain Storage Structures

At Hyattsville, Md., a study on the lowest cost pit-elevator combinations for different grain receiving capacities was completed. A report on the study was prepared and will be published as ARS 52-6, "Selecting Dump Pits and Elevator Legs for Country Grain Elevators."

Research using a system-engineering approach was completed on the layout and design of large country grain elevators of concrete construction. A report entitled "Locating, Designing, and Building Country Grain Elevators" has been prepared and is to be published in the fall of 1965 as an Agricultural Information Bulletin.

### C. Grain Aeration and Drying

1. Drying. At Lafayette, Ind., continuing corn drying studies again showed that dryeration, a combination of high-speed drying and aeration that has been under development for the past two years, not only increases drying capacity but also prevents much of the quality deterioration normally associated with heated-air drying.

Conventional drying and dryeration were compared in tests at three drying air temperatures. Corn was dried from 25 percent to 14 percent moisture in a full-scale continuous-flow dryer. In the conventional drying tests, the hot corn was cooled rapidly in the dryer; with dryeration, the hot corn was removed from the dryer, tempered in a bin for a few hours, and then cooled slowly by aeration. The average hourly drying capacity with dryeration was 197 bushels; with conventional drying, 105 bushels. The drying efficiency averaged 76 percent with dryeration and 58 percent with conventional drying.

From a corn quality standpoint, dryeration reduced the number of stress cracks and the accompanying brittleness of corn that contributes to breakage during handling. With conventional drying, less than 10 percent of the dried kernels were free from stress cracks; with dryeration slightly over 60 percent were without stress cracks.

Two-stage drying, similar to the practice used for drying rice, did not substantially increase drying capacity or efficiency over that obtained with single-stage dryeration. Corn was dried from 25 to about 20 percent moisture, left hot over night in a bin to temper, dried to about 16 percent in a second pass through the dryer, and then allowed to temper a few hours before the drying and cooling were completed by aeration. The additional tempering period with two-stage drying increased the percentage of kernels free of stress cracks from 60 to 72 percent.

The wet milling properties of samples of dried corn, as determined by the prime starch yield test, continued to show that starch yield is dependent on the temperature during drying. With drying air at 190° F., there was no reduction of milling score with any of the drying methods tested. With drying air at 240° F., the milling score was reduced to the lower part of the acceptable range. In the two-stage drying tests, most of the reduction occurred during the first stage. With air at 290° F., the milling score was low in all tests, with damage occurring in both stages of the two-stage tests.



The dryeration process has gained widespread acceptance as a method of drying field-shelled corn and in its first year of application, was used to dry several million bushels of the 1964 crop. Assisted by Purdue University Extension personnel, records were obtained on dryeration systems operated by six farmers and two elevator operators. The cooperators reported increases in drying capacity of from 40 to 80 percent, reduction in fuel and power costs, and cooler corn of better quality than they had experienced with conventional drying. The demand for information on the dryeration process has resulted in articles in nearly all of the leading farm magazines and grain trade journals. Information on the process was presented to about 20 groups in meetings held during the year.

Evaluation of concurrent-flow (air and grain moving in same direction) drying of shelled corn was continued in simulated tests on a digital computer. The mathematical model used to represent the drying process predicted the performance of a concurrent-flow dryer and identified the number and range of the pertinent variables to be investigated in the laboratory.

Eighteen drying tests were performed in a laboratory concurrent-flow dryer developed to investigate the effect of drying air temperatures (up to 400° F.), airflow rate, corn bed depth, and moisture level on dryer performance and corn quality. While drying from 23 percent to 14 percent moisture, the hourly drying capacity ranged from 0.5 bushels per square foot of bed area with 200° F. drying air at 40 c.f.m. per square foot through a 4-foot bed depth, to 2.30 bushels with 400° F. drying air at 97 c.f.m. per square foot through a 2-foot bed depth.

Concurrent drying caused no marked reduction in the brittleness of the corn. There was some reduction in number of stress cracks with a 4-foot corn depth as compared with a 2-foot depth. Most of the drying took place by the time the corn had progressed half-way through the dryer, with an average of slightly under 1 percent moisture removal beyond the half-way point. All the corn dried at 200° F. and that dried from 18 percent moisture at 300° F. showed satisfactory wet milling properties. Corn from all other tests showed borderline or unsatisfactory milling scores. Most of the damage to milling quality occurred during the first half of the drying period when drying temperatures were highest and moisture removal most rapid.

An evaluation of the performance of grain metering devices for grain dryers was strated in the laboratory using a model grain column with clear plastic sides and colored layers of corn. Exploratory tests were conducted with a revolving fluted wheel discharge device similar to that presently used in dryers. A vibratory device was developed that shows promise of providing an unloading method that will minimize grain damage.

At Manhattan, Kans., research to develop and test crossflow ventilation systems for deep silo-type storages was continued in 1964-65. Over the two-year period the full-scale test storage was loaded and emptied 12 times with a total of 300,000 bushels of grain being used in the 12 tests.

In October, 27,000 bushels of new crop grain sorghum averaging over 14 percent moisture (range 12 to 18 percent) and 74° F. were used in a test. After 62 hours of fan ventilation the grain temperature averaged 61° F. and the moisture content 13.6 percent.

In November, a second lot of new crop grain sorghum having an initial average moisture content of 13.4 percent and an average temperature of 73° F. was ventilated. After 64 hours of fan operation the average temperature was 53° F. and the moisture content 12.8 percent.

In December and January a third lot having an initial moisture content of 14 percent and a temperature of 40° F. was used. After 45 hours of ventilation grain temperatures averaged 25° F. and after 112 hours the moisture content averaged 13.6 percent.

A conditioning test was conducted using old wheat. In March, the wheat had an average moisture content of 11.8 percent and a temperature of 45° F. After 40 hours of ventilation grain temperatures averaged 25° F. In April, the grain was warmed to temperatures ranging from 57° to 64° F. with 86 hours of fan operation; the final wheat moisture averaged 12 percent.

Results of the ventilation tests were presented at the Crop Dryer Manufacturers' Council meeting held at Kansas State University in October 1964. The same material was presented in several grain trade publications and in the April 1965 issue of the American Agricultural News Digest.

2. Aeration. At Manhattan, Kans., field studies were continued on a limited basis in commercial storages, both flat and upright.

Three years of test results in a large flat storage, 100 by 360 feet, show the importance and need of proper scheduling of aeration system operation for the first few weeks of the cold weather season. Again during the winter months, serious surface grain crusting, caking, and spoilage developed and covered the center peak area and most of the sloped grain surfaces at depths of 1 to 12 inches. The center peaked grain graded "sample" to a depth of 4 feet with total damage ranging from 12 to 80 percent; and grain moistures increased 6 to 7 percent by January. Aeration fans operated by manual control during February and March completed a cooling stage and reduced grain surface moistures from 17 to 18 percent to 13 to 14 percent. High grain temperatures of 110° F. were reduced to 40° F.

Air temperature and relative humidity limit switches were used to provide automatic control for aerating a test lot of 45,000 bushels of tough wheat in an upright storage. The wheat moisture averaged 13.1 percent, but 10,000 bushels in the lower part of the bin had an average moisture content of 14.4 percent. At an airflow rate of 1/25 c.f.m. per bushel, grain temperatures were reduced to 75° F. in August, to 43° F. in November, and to 32° F. in December. This wheat was loaded into rail cars in June with none of the wheat being downgraded.

Several elevator managers have shown concern about the seemingly excessive weight losses due to moisture evaporation and aeration practices. A reduction in moisture content of 1.0 to 1.2 percent during aeration was considered excessive for several test lots of wheat, because fan operation was extended many hours more than needed to complete a cooling stage or grain was cooled excessively to low temperatures of 20° to 30° F.

A report on changes in Hard Red Winter wheat during storage was completed and submitted for review and clearance for publication. This report summarizes the quality changes for 13 lots during one to two years of aerated storage and 7 lots similarly sampled and stored with aeration. The most significant change for all lots studied was the 10 to 16 percent reduction in sedimentation values, a decrease of about 8 points. Moisture content of the aerated lots of wheat decreased 0.9 percent compared to 0.4 percent for the unaerated lots.

At Hyattsville, Md., data obtained under a research contract on static pressure losses in aeration ducts and through grain near ducts is being analyzed and empirical formulae developed for use in a published report on engineering design of aeration systems.

#### D. Studies on Storing Corn at CCC Bin Sites

At Watseka, Ill., all research was discontinued Dec. 31, 1964. Tests were completed on storing unblended shelled corn in 8 aerated flat storages where fans were operated as blowers on one-half the storages and as exhausters on the others. After two years of storage, with aeration time controlled by a time clock, the average total damage of corn increased 3.7 percentage points when the direction of airflow was up through the corn. Under identical conditions except that the direction of airflow was down, the increase in total damage was 8 percentage points.

Tests were also completed on the effect of various fan operating schedules on maintaining the quality of corn aerated in standard USDA bins. The least increase in total damage occurred when the fan exhausted air from the duct from Sept. to Jan., off from Jan. to March, then blowing into the duct from March until Sept.

Final reports covering the various research studies are now being prepared for ASCS.



## E. Handling, Drying, Aerating, and Storing Rice

1. Drying. At Beaumont, Tex., tests of rice drying procedures using both a pilot-size continuous-flow, heated-air dryer, and laboratory dryer were continued to determine the effect of the saturation deficit (difference between saturated vapor pressure of air at dew point and at dry bulb temperature) of the heated air upon speed of drying, milling yield, and germination of rough rice. Analysis of data from 18 tests in the pilot-size dryer indicates that the speed of drying increased progressively as the saturation deficit of the heated air was increased. For example, drying time was reduced by 35 percent with a saturation deficit of 12 inches of mercury (vapor pressure) as compared with 2 inches. The faster drying was accompanied by some reduction in milling yield of head rice; approximately 3 percent for a saturation deficit of 12 inches of mercury as compared with none at 2 inches. The saturation deficits used had no significant effect on germination. Tests with a laboratory rice dryer produced results similar to those from the pilot-size dryer.
2. Aeration and Storage. At Beaumont, Tex., the series of tests on the use of aeration for maintaining quality of undried rice was continued. Belle Patna rice, initially at 22 percent moisture, was stored in aerated bins for 3, 5, and 6 days and aerated at 0.5, 1.0 and 2.0 c.f.m. per barrel respectively without a reduction in grade; the average ambient temperature was 84° F. during the test period. Nato rice, initially at 19.3 percent moisture was stored for 13, 16, and 17 days and aerated at 0.5, 1.0 and 2.0 c.f.m. per barrel respectively without a reduction in grade; the average ambient temperature was 79° F. TP 49 rice initially at 23.8 percent moisture was stored for 9, 12, and 13 days and aerated at 0.5, 1.0, and 2.0 c.f.m. per barrel without any grade reduction; the average ambient temperature was 66° F. These results will be used in preparing tables or graphs to show "safe" storage procedures for rice, in aerated storage, at various moisture contents and ambient temperatures.
3. Handling of Rough Rice at Commercial Dryers. Studies of handling and operating methods were continued during the 1964 harvest season. At one dryer the four drying units required attention 11 percent of the time. With one operator, there was about 1.6 hours of down time in a 24-hour day due to interference; with two operators the down time was reduced to about 0.3 hour. The four units used 1.8 kilowatt-hours of electricity and 0.19 cubic foot of gas per barrel of rice dried. At a second dryer the four drying units needed attention 15 percent of the time. With one operator down time due to interference was about 3 hours; with two operators down time was reduced to about 0.5 hour. The four units used 2.0 kilowatt-hours of electricity and 0.34 cubic foot of gas per barrel dried. A study was made to determine if handling rice in bucket elevators affected the milling yield of the rice. No significant difference was found in milling yield of samples taken before or after elevating under the conditions of the study.

4. Bulk Handling of Milled Rice. During 1964, research was initiated on handling milled rice in bulk. The first series of tests, with both long- and medium-grain rice, were run to determine the amount of breakage occurring when rice is dropped from various heights onto different surfaces. The milled rice was dropped from heights of 20 and 30 feet onto surfaces of flat and inclined steel, flat and inclined concrete, and on other rice. The most breakage occurred when long-grain rice was dropped 30 feet onto a flat steel surface; the least when short-grain rice was dropped on rice. The height of drop had a significant effect on the amount of breakage. Breakage was reduced when surfaces were inclined 45 degrees.

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COOPERATIVE MARKETING  
Marketing Division, FCS

Problem: Farmers continue to expand their use of cooperatives in marketing the products of their farms. In light of the rapid and complex changes taking place in technology and in market organization and practices, research is needed to help farmer cooperatives and other marketing agencies perform needed marketing services both more efficiently and more effectively. Farmer-directors, managers and others, including the public, need more information to assist in making decisions on how cooperatives can maintain and strengthen the bargaining power of farmers, increase efficiency and reduce costs of marketing, and better meet the needs of our mass distribution system for large quantities of products on a specification basis.

Farmer cooperatives are an important part of the distribution system and represent a major potential for meeting farmers' marketing problems in our modern, dynamic system. They are organized and operated to increase farmers' net income. However, cooperatives face many problems in achieving this goal. Cooperatives must find ways to consolidate volume, for example, through internal growth, merger, acquisition or federation, to strengthen their market position and meet the needs of mass merchandising. Ways must be found to reduce costs by increasing efficiency through improved operating methods, better organization and management, and more use of new technologies.

USDA AND COOPERATIVE PROGRAM

The Department conducts a continuing long-range program of basic and applied research and technical assistance on problems of marketing farm products cooperatively. Studies are made on the organization, operation, and role of farmer cooperatives in marketing. While most of the research is done directly with cooperatives, the results are generally of benefit to other marketing firms. The work is centered in Washington, D. C. Many of the studies, however, are done in cooperation with various State experiment stations, extension services, and departments of agriculture.

The number of Federal professional man-years devoted to this research totals 3.7, of which 2.5 man-years relate to work on grain, 0.3 on rice, 0.8 on feed, and 0.1 to seed.

PROGRAM OF STATE EXPERIMENT STATIONS

Most of the commodity marketing research of the agricultural experiment stations is helpful to marketing cooperatives. Some projects, however, deal specifically with cooperative marketing problems, opportunities, and impacts. At the present time 10 States have 12 research projects in



cooperative marketing. Their commodity distribution is as follows: grain 2, tobacco 1, fruit and vegetables 1, livestock 2, and cross-commodity 6.

Some projects evaluate the performance and organizational features of cooperatives. Different methods of pooling and their problems are studied so as to develop helpful principles. In the analysis of cooperative operations and in working with directors and managers, efforts are made to identify and solve the many problems that are arising. Particular attention is given to what services or functions should be provided by cooperatives. There is interest in learning more about the attitude of members and nonmembers toward cooperative marketing, especially the differences in these attitudes.

In the last few years more attention is being given to the role of cooperatives in achieving bargaining power for farmers. In connection with market structure studies, special attention is being given to the impact of cooperatives on market conduct and performance.

The total research effort on cooperative marketing in the 10 States is 2.5 professional man-years.

#### REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

##### A. Coordination of Marketing

Farmers and their cooperatives need to adapt their marketing methods to the requirements of large-scale buyers, mass merchandising, and other changed conditions. In many cases the coordination of marketing of a number of cooperatives, marketing the produce of hundreds or thousands of farmers is needed to satisfy these needs and improve returns to farmers. Such coordination may be accomplished by establishment of joint sales agencies or by other methods. Research to determine the problems and needs, and to develop guides for adopting new practices, included work with several commodities.

Grain. Changes in freight rates, transportation facilities, and the directions of grain movement call for major changes in grain marketing. A study was made of the economic feasibility of constructing a cooperative terminal export elevator on the Gulf Coast. These regionals now ship around 100 million bushels of grain to the area, which should support an elevator for the benefit of producer-members. A study of cooperative grain marketing and transportation in the Pacific Northwest was completed, including suggestions for cooperative river shipping houses on the Columbia and lower Snake Rivers. Recommendations were made for change in the operations of a multi-purpose local cooperative in Virginia by adding a local elevator and changing some of their grain-buying practices. Continuing is a study for a large regional farm supply cooperative in Tennessee regarding the feasibility of setting up a coordinated grain marketing program, including the operation of local cooperative elevators.

B. Improving Operating and Handling Methods

Research was underway in several commodity fields to examine new methods, equipment, and structures for efficient and safe processing and storage of agricultural products by cooperatives.

Grain. The report on the economics of flat storage at Kansas country elevators was distributed. This was a joint study with Kansas State University.

Rice. Study of drying and storing rough rice on-farm and off-farm continued. Preliminary findings show little differences in quality effects between the stationary and multipass facilities. Commercial dryers tend to perform better on grade, and on-farm dryers on milling quality.

C. Improving the Organization, Financing, and Management of Marketing Cooperatives

Studies were made to determine ways to improve the efficiency and assist cooperatives improve their services by analysis of organization, financing, and management practices.

Grain. Although delayed to some extent, a study is underway on inventory controls, practices, and responsibilities at local cooperative elevators. The annual analysis of the operations and financial status of regional grain cooperatives was continued. These cooperatives are handling increased volumes of grain and with increased emphasis on merchandising, both domestic and export. In view of the rapidly changing transportation situation, several regionals have acquired both water and rail transportation equipment.

D. Improving Sales and Distribution Methods for Farm Production Supplies

Purchasing cooperatives constantly need to be studying ways of increasing volume to lower per unit handling costs, and methods for developing more effective systems of distributing supplies and providing services to rural users.

Feed. A study of 16 local farmer buying groups in Illinois was almost completed. Most groups started by buying feed ingredients for members who had farm mills, but later they began purchasing a number of other farm supplies. The groups had from 11 to 43 members whose farms were twice the size of the average farm in the State, and operators were 10 years younger than the average farmer in Illinois. The principal benefits members realized were: (1) Savings in costs of supplies, (2) ability to obtain some products such as feed additives not readily available before, and (3) better knowledge of ingredients included in livestock rations and more flexibility in changing the composition of rations.

Another study of feed bargaining groups was initiated in California where poultry producers agree on formulas and then bargain with mills to manufacture and deliver the feed to their farms.

A study of the practices of cooperatives in pricing feed at the wholesale and retail level is underway. Special attention is being given to quantity discounts for large versus medium and small purchases of feed and to charges for various services. Field interviews with a number of cooperatives in the eastern half of the United States indicated wide variations in policies and practices. All of the regional wholesale cooperatives in the study allowed their retail outlets some type of price concession based on quantities of specified types of feed. Approximately three-fourths of the retail outlets allowed quantity discounts on individual volumes of feed purchased and two-thirds also gave volume discounts on annual purchases.

#### E. Handling Farm Production Supplies.

Farmers are interested in ways that supplies and services may be provided in the least cost manner and also by methods that will save them time and labor on the farm.

Feed. A study of the use of bulk feed relay stations is underway. Cooperatives in the Northeast added a number of such stations for extending the movement of feed in bulk from their mills during the past year.

Seed. A study of the feasibility of bulk handling of seed is underway. Preliminary findings indicate bulk receiving of seed from farmers is practicable and helpful, but distribution from plants to warehouses has not been feasible with present facilities and equipment.

#### F. Transportation

1. Appraisal of alternative methods of transportation. Field work was completed on a study of ownership and leasing of 202 covered hopper cars operated by six grain marketing cooperatives. All but 40 of the cars were leased. They are used primarily for transporting grain to distant markets.

2. Loss and damage to agricultural products in transit. A study of grain loss and damage in transit via rail and barge was completed. A report of findings, in process, shows grain handlers how to reduce losses by improving transportation and handling equipment and practices.

3. Motortruck operations and costs in hauling feed. Preliminary findings of a study completed on operating costs and practices of 110 bulk feed trucks operated by seven farmer cooperatives include: (1) Total truck operating costs, excluding administrative expenses, averaged 48 cents a



mile operated, (2) operating costs averaged \$2.89 per ton of feed delivered, and (3) direct costs such as drivers' wages and fuel, oil and grease amounted to over three-fourths of total operating costs.

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ECONOMICS OF MARKETING  
Marketing Economics Division, ERS

Problem: Economic research in agricultural marketing revolves around the problems of increasing efficiency in the processing and distribution system and providing a foundation for orderly adjustments to changes inside and outside of agriculture. Marketing must be looked upon as a dynamic and changing process. The capacity to adjust to and cope with the dynamics of modern marketing is required increasingly of producers and distributors of farm products. Demands of a more knowledgeable and sophisticated consuming public are adding to the pressures for an even more rapid escalation of developments and changes within the marketing system. Changes in institutions and redirection of public policies and programs are modifying the economic environment in which marketing firms must perform and operate. Because of rapid changes and increasing complexities associated with a dynamic marketing system, it is necessary that a continuous program of research be conducted in marketing--a program aimed at keeping producers and marketing firms abreast of the flow of events and providing information necessary to them in making proper and orderly adjustments to change.

Of increasing economic concern is the problem of how to improve and strengthen markets for farm products in face of a continuing rise in production, higher distribution costs, and competition from nonagricultural products. The problem of increasing demand for farm products to meet rising productivity has become progressively more pronounced in the last decade. Interest in the development of markets has mounted as larger and larger financial outlays become necessary for price-support operation and maintenance of reasonable levels of farm income.

Through research on the different facets of market development, information is provided farm groups, processors, and distributors which enables them to make the most of new market outlets and opportunities. In addition, they are better able to assess the demand of consumers from the standpoint of kinds and forms of food and services deemed most satisfactory.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing long-term program of economic research relating to grain and forage crops. The research involves economists and personnel with dual economic and technical training and is carried out at Washington, D. C., and field offices in Albany, California; and Peoria, Illinois, and in cooperation with State Agricultural Experiment Stations in a number of States. The grain and forage phases of the research involve 13.1 Federal professional man-years of which 3.0 are studying products and services; 1.0 merchandising and promotion; 0.3 market institutions and market power; 6.6 prices, margins, and costs; and 2.2 locational and interregional competition.

## PROGRAM OF STATE EXPERIMENT STATIONS

### A. Market Institutions and Market Power

Changes in the structure of marketing agricultural products affects the bargaining power of buyers and sellers. These changes also affect marketing practices, services and prices--and ultimately producers, marketing firms and consumers. Research underway at the State stations deals with these changes and some possible alternatives.

Fifteen studies in 13 States are concerned with the direction and magnitude of major changes in firm organization, economic forces, policies, and practices influencing changes in marketing grain and the relationship between economic forces and policies, and the trends in market structure. Information on consolidation, integration, mergers, and their consequences, along with decision-making processes, is being sought. Information on the magnitude and future course of changes would help in expediting and directing feasible trends. Recent extension of operations of many marketing firms toward both sources of supply and product distribution may be materially altering the structure of grain markets. Ease of entrance and exit is also being investigated. Most of the studies concerned with market structure and performance are a part of regional projects. A total of 10.3 professional man-years is devoted to this area of work.

Four hay and feed grain projects are underway at four stations. Three of these projects were part of a western regional project and are concerned with functions performed in marketing hay and feed grains including adequacy of market information on supply, demand and prices, adequacy and acceptability of grade standards, and feasibility of improving market performance. One study of hay marketing in the Southeast is examining the functioning of local markets and with demand projections. These studies involve 2.5 professional man-years at State experiment stations.

### B. Prices, Margins and Costs

One station is studying costs and margins in handling hay in the Pacific Northwest, with emphasis on more efficient alternative methods and possible effects on potential market outlets. State experiment stations' hay marketing research on price, margins, and costs uses .07 of a man-year.

Five States are conducting studies concerned with supply, demand, utilization, and pricing of wheat. The question of demand for specific grains and qualities and how accurately local prices reflect demand is being studied on a limited basis. Expansion of market outlets, new uses and changes in utilization patterns, and supply and flow patterns of wheat having specific quality characteristics are being studied by three States.



Closely related to price studies are such items as costs, margins, and efficiency of operation and firm management. A total of 6 professional man-years is involved.

### C. Location and Interregional Competition

Changes in the technology of producing and processing agricultural products has much impact on the comparative advantage of one producing area over another. Changes in transportation rates and costs also affect the competitive position of some areas more than others. These changes and their effects on the location and interregional competition are being studied by the State stations.

Since grain marketing and processing facilities require extensive capital investments and can be used for few other purposes, correct decisions relative to type and location are important. Sixteen studies concerned with transportation and location of grain marketing facilities are underway. Most of these studies contribute to regional undertakings. The researchers in the north central and southern regions have exchanged information and are coordinating their research to maximize the efforts of both regions. Some of the work in this area is closely related to the research reported under Market Institutions and Market Power.

Transportation rates, services, and relative prices undergo changes that cause shifts in the source and destination of grains and in the type of facilities needed for handling grains. The changes in freight rates in recent years are having significant effects, especially in the southern region. The impact of increased exports also has had its effect on location of needed facilities in the North Central States. A total of 14.3 professional man-years is involved.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Prices, Margins, and Costs

Trends in marketing spreads for grain and grain products were mixed during 1964-65 while the farm value of these commodities remained largely unchanged from the level of the past several years. The retail price of a 1-pound loaf of white bread increased during the second half of 1964 from 20.6 to 21 cents; during the first half of 1965 it fluctuated from 20.9 to 21 cents. The wholesaler-baker spread continued to account for more than one-half the retail bread price and averaged 11.4 cents a loaf in 1963 and 1964. Increases in wages, salaries, and fringe benefits, which now account for more than half of the wholesaler-baker spread, were partially offset by lower packaging and wrapping costs.

Dry cereal prices have increased steadily while costs of corn and other raw materials remained virtually unchanged. The retail price of a 12-ounce package of corn flakes increased from 16.9 cents in 1947-49 to 28.6 cents in 1964 and to 28.9 cents in recent months. The farm value was 2.5 cents in 1964, compared with 2.6 cents in 1963 and an average of 2.7 cents during 1947-49.

The cost of price support programs for grains is materially affected by the handling and storage rates paid by the U.S. Department of Agriculture. Although rates paid by the Department are periodically reviewed, negotiated and adjusted in an attempt to reflect changing cost structures, rates paid by the Commodity Credit Corporation have been fixed without precise and definite information as to actual costs of providing these services. Two administrative reports have been submitted to the Agricultural Stabilization and Conservation Service regarding the cost of handling grains in commercial elevators. Cost comparisons were made by mode of transportation, type of plant and kind of grains handled. Work is continuing and will include costs for storage as well as handling, taking into account regional differences, capacities utilized, grains stored, transportation costs and type of construction. Administrative reports will be prepared and submitted to the Agricultural Stabilization and Conservation Service. A study conducted under contract by Montana State University showed that elevators in the spring wheat area, geared to World War II marketing patterns, were forced to expand rapidly after the War due to a shortened harvest period and increased production. Five elevator models were developed and the cost of handling grain ranged from 0.037 cent per bushel for those with an annual volume of 1.75 million bushels to 0.109 cent for those which handled only 75,000 bushels. One report gives fixed and variable cost including those for nongrain activities. A second report projects elevator numbers, sizes, locations, and functions to 1980.

Grain banks have become an integral part of the mixed feeds industry in the Midwest with potential savings for both the farmer and the feed mill. It is estimated that use of grain banks can increase output of a typical plant by 75 percent without additional labor. Expansion and use of grain banking has been encouraged by the development of mechanical bulk handling equipment at the farm and the plant. Costs and charges for grain banks varied widely, but the average cost of processing feed was \$8.95 per ton. Another important change taking place has been in the actual manufacturing operation. Companies have decentralized production facilities. Smaller and more efficient facilities have been established in consumption areas. A series of surveys has been conducted in recent years in an attempt to provide management with economic engineering model data on labor and capital requirements. The most recent study on processing ingredients points out that this is basically a machine operation and that a minimum amount of labor is required once the grinding, crimping, or corncracking equipment has been

started. A small model processing 45 tons of ingredients per 8-hour day would require 0.07 man-hour per ton while a larger model processing 120 tons a day would require only 0.03 man-hour per ton. Operating costs per ton for the small model are 40 percent greater than the costs for larger models. Data from all surveys are being compiled into a general report which will analyze costs for the entire manufacturing operation with varying degrees of product specialization.

Operating practices and labor utilization in rice mills were studied. Results indicate that if the rice milling industry were to use the most efficient methods observed and attain the labor work standards described, there is a potential production labor savings of about 50 percent. Such a saving means nearly 10 cents per 100 pounds of rough rice milled or some \$7 million annually.

Present hay grades do not reflect all the quality attributes important in pricing, and trading is disorganized. Data on selected alfalfa hay quality factors were obtained at various levels of marketing in Arizona, California, Nevada, and Oregon. Based on these data, experimental grades were developed along with the approximate cost of applying these grades to individual lots of hay. In addition, the experimental grades were market tested to determine their acceptability.

#### B. Location and Interregional Competition

The grain marketing system is currently faced with problems of substantial change. Rapid increases in volumes of whole grain flowing through the gulf ports for export have resulted in construction of new terminal elevator facilities near production areas. Other facilities are proposed. The flour milling industry is in the process of relocating away from the production areas because of changes in transportation rates causing changes throughout the grain markets including millfeed users.

Research on the location and performance of the whole grain industry in the Northeast, conducted in cooperation with the Maryland Agricultural Experiment Station, has been completed and a report published. Comparable studies are underway in the rest of the United States including one under contract with Oklahoma State University. When these studies are completed, data will be available to evaluate present and probable interregional competition in the U.S. grain markets. Results to date indicate increasing off-farm sales, decreasing numbers of grain merchandisers and processors, shifts in the spatial location of facilities, increasing integration, and increasing exports. Current industry problems appear to be centered largely around transportation methods and rates.



In the Northeast, for which data are complete, there are few strictly regional grain interests. Except for volumes moving through integrated establishments with grain interests outside the region, the volume of grain moving into the Northeast has declined sharply. The number of establishments which have gone out of business was as high as 20 percent in some segments of the industry and a preponderance of these were independent nonintegrated establishments.

Rail transportation of grains shipped by country elevators in the north central region decreased relatively by more than 10 percent in a 5-year period ending in 1963. Barge and truck shares increased accordingly. These shifts led to rate reductions by railroads in attempts to regain traffic. Such changes already have caused some adjustments in the location of storage and milling operations, and may well cause many more in the near future.

The pattern of shifting grain transportation from rail to truck also occurred in the Northwestern States. It was found that considerable shipments now bypass country elevators, moving directly from farms to terminal markets. This type of movement results in a change of market structure and the nature of competition in the markets in which farmers sell grain. The shift from rail to truck transportation was due to changes in relative rates rather than changes in service factors associated with the different modes of transport.

Food and feed grain exporters must be careful in timing their demands for shipping, according to an analysis of trampship charter rates for the period 1961-1965. These rates fluctuate widely, and these fluctuations seemed to respond to causes outside the grain trade. The study also noted that trampship rates are considerably affected by country of registry, with U.S. flag vessels having rates well above their foreign registry counterpart vessels.

### C. Products and Services

Another special area receiving increased emphasis is cereal grains. The need for market potentials research is vital because grain products are especially vulnerable to market substitution. The emphasis in recent years has been to depict the precise nature of the technical and economic competition facing these agricultural materials so as to provide guidelines on the cost-price relationship and performance characteristics that are needed to resist further market erosion. The emphasis has been on researching starches. The efficient utilization of starch, the major component of cereal grains, is the key to expanded use of cereal products. Work also began on market potentials for frozen doughs and batters for household use, as well as for point-of-sale defrosting and bake-off. This study is an outgrowth of USDA's previous research on freezing practices in the baking

industry. The new study will explore a growing new industry which uses freezing for unbaked dough and batter. This practice may have potential for increasing consumption and lowering prices to consumers for bread.

#### D. Merchandising and Promotion

The feasibility and value of using computers to simulate information required in agricultural marketing firms' decision making has been illustrated by research conducted with feed manufacturing and retail food firms. A study of mixed feed operations showed that linear programming can be a valuable management tool for computing feed formulas, anticipating changes in ingredient usage rates, and developing feed specifications. Gross savings of \$1.70 per ton were attributed to the use of LP during a 23-week test period in two firms. Linear programming appears to be profitable for the small- and medium-sized manufacturers as well as the larger ones. Also, similar techniques can be employed by management to assemble cost information on procurement, inventory, ingredients, transportation, and distribution, and to arrive at a system of distribution, plant location, plant size, transportation facilities, inventory of drugs, etc., that will provide farmers with lower cost feeds. Management techniques now being tested for an entire feed manufacturing firm operation should apply to other agricultural industries.

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CONSUMER PREFERENCE AND QUALITY DISCRIMINATION--  
HOUSEHOLD AND INDUSTRIAL  
Standards and Research Division, SRS

Problem. With the increasing complexity of marketing channels and methods, it has become almost impossible for consumers to express to producers either pleasure or displeasure with available merchandise. To market agricultural products more effectively, it is necessary to understand existing household, institutional, and industrial markets and the reasons behind consumers' decisions to purchase or not to purchase. Information is needed on consumers' attitudes toward old and new product forms of agricultural commodities, preferences, levels of information or misinformation, satisfactions or dislikes, and what product characteristics would better satisfy current consumers and/or attract new ones. It is also important to know the relationship between the consumption of one agricultural commodity and another in consumers' patterns of use, the relationship between agricultural and nonagricultural products, and probable trends in the consumption of farm products. Producer and industry groups as well as marketing agencies consider such information essential in planning programs to maintain and expand markets for agricultural commodities which, in turn, increases returns to growers.

USDA AND COOPERATIVE PROGRAM

The Special Surveys Branch conducts applied research among representative samples of industrial, institutional, or household consumers and potential consumers. Such research may be conducted to determine preferences, opinions, buying practices, and use habits with respect to various agricultural commodities; the role of competitive products; acceptance of new or improved products; and consumers' ability to discriminate among selected attributes of a product or levels of an attribute, and the preferences associated with discriminable forms.

In addition to the studies of consumer preference and discrimination, the Branch also provides consultants and conducts special studies, upon request, for other agencies in the USDA or within the Federal Government, when survey methods can be usefully applied to the evaluation of programs, services, or regulatory procedures of interest to the requesting agencies.

The research is carried out in cooperation with other USDA or Federal agencies, State experiment stations, departments of agriculture, and land-grant colleges, and agricultural producer, processor, and distributor groups. Closely supervised contracts with private research firms are used for nationwide surveys; studies in selected areas are usually conducted by the Washington staff with the assistance of locally recruited personnel.

The Branch maintains all of its research scientists, who are trained in social psychology or other social sciences, in Washington, D. C., which is headquarters for all the research whether it is conducted under contract or directly by the Branch. Work on consumer preference for cereals involves 2.3 Federal professional man-years.

#### REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

##### Consumer Preference for Cereals

A contract has been signed with a private research firm to collect information from a nationwide sample of homemakers on their use of and opinions about selected potato, rice, and wheat products. The study is designed to ascertain household consumers' preferences and buying practices for these commodities, including specialty or highly processed convenience-type food products, and to identify the characteristics which consumers consider to be important in selecting such food products and to determine their satisfaction with items currently available. The field work on this study is planned for the fall of 1965.

#### PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

None

ECONOMIC AND STATISTICAL ANALYSIS  
Economics and Statistical Analysis Division, ERS

Problem. Adequate and accurate information is needed on supplies, production and consumption of farm products, and the effects these and other factors have on the prices of agricultural commodities. Such information is needed in planning operations for the producers, processors and distributors and also benefits the consumer in selecting his purchases. Similarly accurate quantitative knowledge of the interrelationships among prices, production and consumption of farm products are needed by Congress and the Administrators of farm programs to effectively evaluate current and future price support and production control programs.

Due to the instability of the prices he receives, the farmer stands in special need of accurate appraisals of his economic prospects if he is to plan and carry out his production and marketing activities in an efficient and profitable way. The farmer needs to be provided with economic facts and interpretations comparable to those available to business and industry, through a continuous flow of current outlook intelligence and the development of longer range projections of the economic prospects for the principal agricultural commodities.

USDA AND COOPERATIVE PROGRAM

The Department has a continuing program of basic research concerning the factors affecting prices, supply, and consumption of principal agricultural commodities and the analyses of the situation and outlook for selected commodities. The Federal professional man-years involved for grain are 5.5 annually, of which 1.5 are devoted to work on the supply and demand for grains and 4.0 to work on the grain situation and outlook. The program is carried on in Washington, D. C.

PROGRAM OF STATE EXPERIMENT STATIONS

For the most part the States depend on the U. S. Department of Agriculture for the yearly cross-the-board commodity situation and outlook research. There is increasing interest in longer range price prediction because of the growing specialization of farms, which makes yearly enterprise shifts less common and less feasible, and which calls for large capital commitments over longer periods of time. The State extension staff members supplement and adapt such research information to meet the commodity situation of their States.

The total direct research effort in the situation and outlook area is small--probably no more than 2 to 3 professional man-years. While not designated as outlook research, much of the research conducted by the



experiment stations and reported elsewhere contributes to improved understanding of price-making forces, which in turn improves market situation analysis and price forecasting.

Many of the States carry on supply, demand, and price analyses for the products of their State. Much of the research is commodity oriented, though some projects are of a highly mathematical and theoretical nature aimed at improving price analysis methodology.

The research on demand for a large number of commodities will indicate the price elasticity, the income elasticity, and the cross elasticities of the commodities being studied. Because researchers are finding that some changes cannot be explained by price, income, and supply of competing commodities there is increasing research interest in social and psychological factors affecting demand.

The supply response to price changes is a matter that is receiving considerable attention. This is in part because of its significance to farm incomes and government programs. Significant progress is being made in understanding the relationship of the capital structure on farms to supply response and thus to the difference between long-run and short-run supply responses.

## PROGRESS -- USDA AND COOPERATIVE PROGRAMS

### A. Demand, Supply and Price Analysis

1. Food Grains. A special study was made of the cash market for soft red winter wheat at Chicago at the request of the Commodity Exchange Authority. This study identified and measured the influence of basic supply and demand factors affecting the season average price and the May average price for soft red winter wheat. The results of this study will be useful in an analysis of the supply and demand factors for the other classes of wheat.

A study was made for the Secretary's Office of the demand for rice and the effect of different levels of prices on consumption of rice. This study concluded that consumption was not very responsive to price changes--i.e., an increase of 10 percent in the price of rice could result in a decrease of about 2 to 3 percent in rice consumption. The study also found that per capita demand for rice had increased in recent years. However, because of lack of data, it was not possible to determine the extent new products such as instant rice and new consumer attitudes concerning the place of rice in our diet contributed to this rise in consumption.

## 2. Feed Grains and Byproduct Feeds

Analyses were made of the seasonal variations of price of 4 feed grains and 17 byproduct feeds and the results were published in the June and August issues of the Feed Situation.

With the growing importance of soybeans in the feed and fats and oils economy, some work was initiated on the demand for soybean meal and soybean oil. Several demand functions for soybean meal and soybean oil have been statistically tested. Preliminary results from this analysis indicate that, on the average, a 10-percent increase in the price of soybean meal will result in a decrease in consumption of soybean meal by about 6 percent. On the other hand, a 10-percent decrease in the price of cottonseed meal, its nearest competitor, will only decrease consumption of soybean meal by about 3 percent or only about half of the effect of a like increase in soybean meal prices. These are the effects of prices after the quantity of feed grains utilized has been taken into account.

### B. Situation and Outlook Analysis

#### 1. Food Grains

Wheat. During the past year special attention was again devoted to analysis of proposed wheat programs. Program analysis was directed at the alternatives resulting from the substitution of wheat and feed grain acreages and the effect on production and supplies. Under the existing wheat program more wheat has been fed. Coverage of the Wheat Situation was expanded to devote more attention to the competitive position of wheat and feed grains. Another new feature was the development of statistical data on wheat and flour price relationships and the factors affecting them. These materials are updated and discussed periodically. A special analysis was prepared on trends in wheat production and results were published in July 1965. There were some significant changes in production of the various classes. Production of both hard winter wheat and white wheat increased substantially and account for larger proportions of the crop in recent years than in the past. Production of the other classes, hard spring, soft red winter and durum, has generally diminished, although production of the last two continues to fluctuate sharply from year to year.

Rice. Statistics covering supply and disappearance of milled rice were expanded and historical data were revised to account for the increasing trend to brown and parboil rice. Special attention is also being given to competition of foreign rice in export markets.

## 2. Feed

During 1964-65, the Feed Grain Program continued to be given special attention. The program was a dominant factor in the smaller feed grain acreage in 1961-65, about 20 percent below the 1959-60 base, a reduction in carry-over stocks from 85 million tons in 1961 to 54 million this year, and an increase of 16 percent in feed grain prices since 1960-61. The marked upward trend in yield per acre, averaging about 6 percent annually since 1954, brought each of the 4 feed grain yields to new record highs in 1965 and resulted in another big crop in spite of the lowest acreage in more than 70 years. Special analyses were made, in cooperation with other agencies of the Department, of alternative types of feed grain programs under consideration for 1966 and later years.

A special study was made of the relationship between livestock and feed prices, feeding rates per animal unit and total feed grain consumption during the past 10 years. Results of the study were discussed and shown graphically in the November 1964 issue of the Feed Situation.

An analysis of the seasonal variation in the production of commercially prepared livestock and poultry feeds, based on data furnished by the American Feed Manufacturers Association, was published in the February 1965 issue of the Situation. High-protein feed supplies, consumption per animal unit, and prices continued to be given special attention, as were changes in the relationship between prices of urea-grain mixtures and oilseed meals.

### PUBLICATIONS -- USDA AND COOPERATIVE PROGRAMS

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- Askew, W. R. Rice Situation. Published annually. ERS, USDA, Washington, D.C.
- Clough, Malcolm. Feed Situation. Published 5 times a year. ERS, USDA, Washington, D. C.
- Clough, Malcolm. November 1964. Recent trends in feed consumption and livestock-feed relationships. Feed Situation, pp. 40-44.
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- Gomme, F. R. July 1965. Trends in U.S. wheat production by classes. Wheat Situation, pp. 16-17.
- Grain and Feed Statistics. March 1965. Supplement for 1964 to Statistical Bulletin No. 159, 65 pp.
- Ross, J. S. June 1965. Seasonal Variation in Feed Grain Prices. Feed Situation, pp. 28-33.
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